



**KIZILÖTESİ UYGULAMASININ ÇİYA TOHUMUNUN  
BAZI BİLEŞENLERİ, FONKSİYONEL ÖZELLİKLERİ VE  
MÜSİLAJ ÖZELLİKLERİ ÜZERİNE ETKİSİ**

**EFFECTS OF INFRARED TREATMENT ON SOME  
CONSTITUENTS, FUNCTIONAL PROPERTIES AND  
MUCILAGE PROPERTIES OF CHIA SEED**

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*To my little cousin Nisa..*



## **ABSTRACT**

# **EFFECTS OF INFRARED TREATMENT ON SOME CONSTITUENTS, FUNCTIONAL PROPERTIES AND MUCILAGE PROPERTIES OF CHIA SEED**

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Chia seed (*Salvia hispanica*) has high nutritional value and positive effect on health due to its protein and total dietary fiber contents, phenolic compounds, and essential oils. Mucilage extracted from chia can be used as a gelling agent, foaming agent, emulsifier, and coating material in food industry.

In recent years, utilization of infrared (IR) treatment in food industry is increasing, due to its advantages (direct heat penetration, fast heating rate, short processing time, energy saving, low cost) over conventional heating. Infrared treatment has been used for drying (fruits, vegetables, grains etc.), cooking, roasting, thawing, sterilization, enzyme inactivation, and for enhancing the extraction of some bioactive compounds (polyphenols, flavonoids etc.).

In literature, microwave, roasting, and autoclaving have been applied to chia seeds for various purposes such as extracting health beneficial components, increasing extraction efficiency or improving functional, and organoleptic properties and storage stability.

To the best of our knowledge, in literature effects of infrared treatment on some constituents, functional properties, and mucilage properties of chia seed were not investigated. Therefore, in this study, chia seeds were infrared treated at different powers (700W, 900W and 1100W) and times (25 and 50min). The effects of infrared treatment on protein, ash, color, total dietary fiber, total phenolic content, total flavonoid content,

phenolic profile (by HPLC), antioxidant activity (DPPH, TAC), FTIR spectrum, and functional properties (water and oil holding capacity, emulsion activity, and stability) of chia samples were investigated. Mucilage was extracted from control and infrared treated chia samples and mucilage properties were determined. The effects of infrared treatment on the yield, protein, ash, color, total dietary fiber, functional properties (water and oil holding capacity, emulsion activity, and stability, viscosity) and FTIR spectrum of mucilage samples were investigated.

In the present study, infrared treatment caused increases in total phenolic content, total flavonoid content (except 1100W), antioxidant activity, chlorogenic acid, rutin, ferulic acid (except 1100W-50 min), quercetin (except 700W-25 min, 900W-50 min and 1100W), emulsion activity (except 900W-50 min and 1100W), emulsion stability (except 900W and 1100W), and total dietary fiber (except 1100W) of chia samples. Infrared treatment of chia samples resulted in significant increases in yield, emulsion activity (at 700W and 900W), and emulsion stability (at 700W and 900W) of mucilage samples. Infrared treatment of chia samples at all powers and times caused a negative effect on water and oil holding capacities of chia and mucilage samples. FTIR spectrums for infrared treated chia samples and mucilage samples extracted from these chia samples have all the specific peaks reported in literature. Infrared treatment of chia samples only at 900W-50min and 1100W-50min resulted in lower viscosity values for mucilage samples (1% and 2%, w/v).

For food industry, utilization of infrared treated chia is promising as a high value raw material due to its high amount of health beneficial constituents. Mucilage extracted from chia seeds takes part in many industrial applications (food, chemistry, etc.). Infrared treatment of chia seeds is also promising in terms of increased mucilage yield and improved some mucilage properties.

**Keywords:** Chia, Infrared Treatment, Mucilage Extraction, Mucilage Yield, Phenolic Profile, Antioxidant Activity, Total Dietary Fiber, Functional Properties, FTIR

## ÖZET

# KIZILÖTESİ UYGULAMASININ ÇİYA TOHUMUNUN BAZI BİLEŞENLERİ, FONKSİYONEL ÖZELLİKLERİ VE MÜSİLAJ ÖZELLİKLERİ ÜZERİNE ETKİSİ

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Çiya tohumu (*Salvia hispanica*), yüksek besleyici değere sahiptir ve içerdiği protein, toplam besinsel lif, fenolik bileşenler ve esansiyel yağlardan dolayı sağlık üzerine olumlu etkisi vardır. Çiyadan ekstrakte edilen müsilaj, gıda endüstrisinde jelleştirme ajanı, köpük oluşturucu ajan, emülgatör ve kaplama materyali olarak kullanılabilir.

Son yıllarda, kızılötesi (IR; infrared) uygulamasının gıda endüstrisinde kullanımı konvansiyonel ısıtmaya kıyasla avantajlarından (doğrudan ürüne penetrasyon, hızlı ısıtma, kısa işlem süresi, enerji tasarrufu, düşük maliyet) dolayı artmaktadır. Kızılötesi uygulaması, kurutma (meyve, sebze, tahıl vb.), pişirme, kavurma, çözündürme, sterilizasyon, enzim inaktivasyonu ve biyoaktif bileşiklerin (polifenoller, flavonoidler vb.) ekstraksiyonunu arttırmak için kullanılmaktadır.

Literatürde, çiya tohumlarında sağlık açısından yararlı bileşenlerin ekstraksiyonu, ekstraksiyon verimliliğinin artırılması, fonksiyonel ve organoleptik özelliklerinin ve depolama stabilitesinin iyileştirilmesi gibi farklı amaçlar için mikrodalga, kavurma ve otoklavlama uygulanmaktadır.

Bilgimiz dahilinde, literatürde kızılötesi uygulamasının çiya tohumunun bazı bileşenleri, fonksiyonel özellikleri ve müsilaj özellikleri üzerine etkisi çalışılmamıştır. Bundan dolayı, bu çalışmada çiya tohumlarına farklı güçlerde (700W, 900W ve 1100W) ve sürelerde (25 ve 50 dk) kızılötesi uygulanmıştır. Kızılötesi uygulamasının, çiya

örneklerinin, protein, kül, renk, toplam besinsel lif, toplam fenolik madde içeriği, toplam flavonoid madde içeriği, fenolik profil (HPLC ile), antioksidan aktivite (DPPH, TAC), FTIR spektrumu ve fonksiyonel özellikler (su ve yağ tutma kapasitesi, emülsiyon aktivitesi ve stabilitesi) üzerine etkisi incelenmiştir. Müsilaj, kontrol ve kızılötesi uygulanmış çiya örneklerinden ekstrakte edilmiş ve müsilaj özellikleri belirlenmiştir. Kızılötesi uygulamasının müsilaj örneklerinin verim, protein, kül, renk, toplam besinsel lif, fonksiyonel özellikler (su ve yağ tutma kapasitesi, emülsiyon aktivitesi ve stabilitesi, viskozite) ve FTIR spektrumu üzerine etkisi incelenmiştir.

Bu çalışmada, kızılötesi uygulaması ile çiya örneklerinin, toplam fenolik madde miktarı, toplam flavonoid madde miktarı (1100W hariç), antioksidan aktivite, klorojenik asit, rutin, ferulik asit (1100W-50 dk hariç), kuersetin (700W-25 dk, 900W-50 dk ve 1100W hariç), emülsiyon aktivitesi (900W-50 min ve 1100W hariç), emülsiyon stabilitesi (900W ve 1100W hariç) ve toplam besinsel lif miktarı (1100W hariç) artmıştır. Çiya örneklerine kızılötesi uygulaması, müsilaj örneklerinin veriminde, emülsiyon aktivitesinde (700W ve 900W'da) ve emülsiyon stabilitesinde (700W ve 900W'da) istatistiksel olarak önemli artışlara yol açmıştır. Çiya örneklerine tüm güç ve sürelerde kızılötesi uygulaması, çiya ve müsilaj örneklerinin su ve yağ tutma kapasitesi üzerinde olumsuz etkiye neden olmuştur. Kızılötesi uygulanan çiya örnekleri ve bu çiya örneklerinden elde edilen müsilaj örneklerinin FTIR spektrumları, literatürde belirtilen tüm spesifik pikleri içermektedir. Çiyaya sadece 900W-50dk ve 1100W-50dk kızılötesi uygulaması ile müsilaj örneklerinde (%1 ve %2, w/v) daha düşük viskozite değerleri elde edilmiştir.

Gıda endüstrisinde, kızılötesi uygulanmış çiyanın, yüksek miktardaki sağlığa yararlı bileşenlerinden dolayı değerli bir ham madde olarak kullanımı umut vericidir. Çiya tohumlarından elde edilen müsilaj birçok endüstriyel uygulamada (gıda, kimya, vb.) yer almaktadır. Çiya tohumlarına kızılötesi uygulaması müsilaj veriminin artması ve müsilaj özelliklerinin iyileştirilmesi açısından da ayrıca umut vericidir.

**Anahtar Kelimeler:** Çiya, Kızılötesi Uygulaması, Müsilaj Ekstraksiyonu, Müsilaj Verimi, Fenolik Profil, Antioksidan Aktivitesi, Toplam Besinsel Lif, Fonksiyonel Özellikler, FTIR



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## **SYMBOLS AND ABBREVIATIONS**

### **Symbols**

dw                      Dry weight

### **Abbreviations**

AACC                      American Association of Cereal Chemists

AOAC                      Association of Official Analytical Chemists

ATR-FTIR                      Attenuated Total Reflectance-Fourier Transform Infrared Spectroscopy

ANOVA                      Analysis of Variance

CE                          Catechin Equivalent

DPPH                      2,2-Diphenyl-1-picrylhydrazyl

FIR                          Far Infrared

GAE                          Gallic Acid Equivalent

IR                              Infrared

MWD                      Multiple Wavelength Detector

TAC                          Total Antioxidant Capacity

TE                              Trolox Equivalent

QE                              Quercetin





# 1. INTRODUCTION

Chia (*Salvia hispanica*) is a pseudocereal with small seed and oval shape. It has a dark brown to beige color with small dark spots on it. Chia seed has high nutritional value and positive impact on health (antioxidant activity, prevents cardiovascular diseases, and certain types of cancers etc.) due to its higher phenolic compounds, essential oils, protein, and total dietary fiber content (Coelho and Salas-Mellado, 2014). Chia seeds have functional properties such as high water and oil holding capacity and emulsion activity.

Chia seeds contain 5-7% mucilage (Fernandes et al., 2020; Capitani et al., 2012; Hernández, 2012). Chia mucilage is a water soluble, complex polysaccharide with high molecular weight and viscosity. Mucilage can be used as a gelling agent, foaming agent, emulsifier and coating material in food industry due to its functional properties such as water and oil holding capacity and emulsion activity (Reyes-Caudillo et al., 2008). In literature, for mucilage extraction, conventional heat treatment, sonication and microwave was used (Hernández, 2012; Nayani, 2020; Urbizo-Reyes et al., 2019).

Infrared treatment (IR) has become increasingly popular in food industry due to its advantages over conventional heating, such as direct heat penetration, energy saving, short processing time, and uniform temperature distribution (Krishnamurthy et al., 2008). Infrared power and treatment time affect drying kinetics and quality of food (Huang et al., 2021). Infrared has been used for drying (fruits, vegetables, grains, etc.) cooking, baking, thawing, roasting, boiling, pasteurization, sterilization, blanching, and inactivation of enzymes (Rastogi, 2021). Infrared was used in the production of noodle, bulgur, heat-moisture treated starch, bread, cookie, cake and cracker (Basman and Yalçın, 2011; Savas and Basman, 2015; Sumnu and Keskin, 2010). Infrared was also used for inactivation of enzymes (lipoxygenase, urease, polyphenoloxidase, lipase, phosphatase, etc.) (Yalcin and Basman, 2015; Lin et al., 2009). Utilization of infrared facilitates extraction of many food components (polyphenols, flavonoids, etc.) by disintegrating the structure and results in higher extraction yield (Xiang et al., 2022). Microwave, roasting and autoclaving have also been used for extracting health beneficial components, increasing extraction efficiency or improving functional and organoleptic properties and storage stability (Xiang et al., 2022).

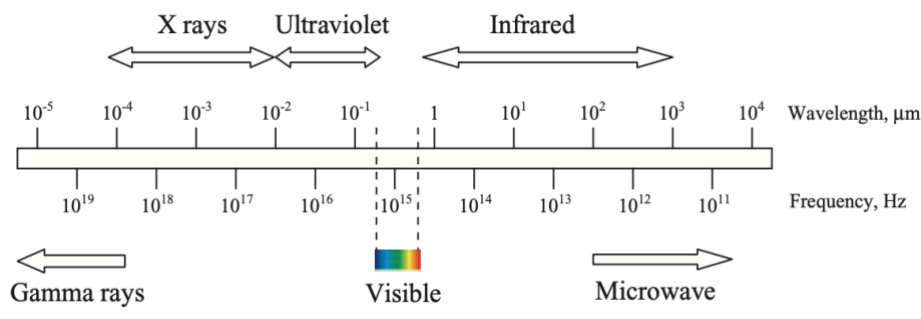
To the best of our knowledge, in literature there is no study about the effects of infrared treatment on some constituents, functional properties and mucilage properties of chia seed. Therefore, in the first part of the study, chia seeds were infrared treated at different powers (700, 900W, and 1100W) and times (25 and 50 min). The effects of infrared treatment on protein, ash, color, total phenolic content, total flavonoid content, phenolic profile (by HPLC), antioxidant activity (DPPH, TAC), total dietary fiber, functional properties (water and oil holding capacity, emulsion activity, emulsion stability), and FTIR spectrums of chia samples were investigated. In the second part of the study, mucilage was extracted from control and infrared treated chia samples. The effects of infrared treatment on the yield, protein, ash, color, total dietary fiber, functional properties (water and oil holding capacity, emulsion activity, emulsion stability, viscosity) and FTIR spectrum of mucilage samples were investigated.

## 2. GENERAL INFORMATION

### 2.1. Infrared

In recent years, utilization of infrared (IR; Infrared) treatment in food industry is increasing, due to its advantages (efficient, direct heat penetration, fast heating rate, short processing time, energy saving, low cost) over conventional heating (Rastogi, 2021). Infrared is transferred directly from the source to product surface without heating the surrounding air (Sakare et al., 2020).

IR radiation is a type of energy that is transferred by electromagnetic waves. IR radiation is between the spectrum of visible light (0.38- 0.78  $\mu\text{m}$ ) and microwave (1-1000 mm) (as shown in Figure 1). IR can be divided into three regions based on their wavelengths: near infrared (0.75-1.4  $\mu\text{m}$ ), mid infrared (1.4-3  $\mu\text{m}$ ), and far infrared (3-1000  $\mu\text{m}$ ) (Rastogi, 2021). The penetration ability of short waves is ten times greater than that of long waves. The direct penetration capability of infrared radiation increase energy flux without surface burning, thereby reduces the required heating time as compared to that of conventional heating methods. This situation is valid for thin products (Skjöldebrand, 2002).



**Figure 1.** Electromagnetic wave spectrum (Krishnamurthy et al., 2008)

The interaction of food with infrared radiation leads to two possible outcomes: reflection and absorption. The absorbed part of the infrared radiation causes stretching, vibrational, and rotational motion. This affects the physicochemical, textural and rheological properties of the food (Aboud et al., 2019) (Krishnamurthy et al., 2008).

The absorption intensities at different wavelengths varies depending on the food components (Krishnamurthy et al., 2008). Food consists of various organic and inorganic compounds. Amino acids, polypeptides and proteins exhibit two strong absorption bands

at 3-4 $\mu\text{m}$  and 6-9 $\mu\text{m}$  wavelengths and carbohydrates show two strong absorption bands at 3 $\mu\text{m}$ , 7-10 $\mu\text{m}$  while lipids have three strong absorption bands at 3-4 $\mu\text{m}$ , 6 $\mu\text{m}$ , and 9-10 $\mu\text{m}$  wavelengths (Sandu, 1986).

The composition and structure of the food and IR wavelength determine the penetration depth of the infrared in the food. When NIR (0.75 to 1.4 $\mu\text{m}$ ) is used, the penetration depth is 1.5 mm for carrot, 2 mm for wheat grain, 4-6 mm for biscuit, and 11-12 mm for bread (Krishnamurthy et al., 2008).

IR has been used for drying (fruits, vegetables, grains, etc.) cooking, baking, thawing, roasting, boiling, pasteurization, sterilization, blanching, and inactivation of enzymes (Rastogi, 2021). Infrared was used in the production of noodle, bulgur, heat-moisture treated starch, bread, cookie, cake, and cracker (Basman and Yalçın, 2011; Savas and Basman, 2015; Sumnu and Keskin, 2010). Infrared was also used for inactivation of enzymes (lipoxygenase, urease, polyphenoloxidase, lipase, phosphatase, etc.) (Aboud et al., 2019; Yalcin and Basman, 2015; Lin et al., 2009).

Infrared power and treatment time affect drying kinetics and quality of food. Barley, rice, apple, carrot, red pepper, potato, corn, tomato, blueberries, and onions were dried successfully by using infrared (Tuncel, 2016; Huang et al., 2021). However, limited penetration depth and potential breakage and cracking of materials such as barley and rice during prolonged processes are some of the disadvantages (Krishnamurthy et al., 2008).

In a study by Yilmaz and Tuncel (2010), infrared, hot air drying, and infrared-hot air drying was used to reduce the moisture content of corn kernels from 29%, 24%, 18.5%, 16%, and 15% to 13%. It was reported that energy expenses for reducing the moisture content from 15% to 13% by using hot air drying were higher as compared to the expenses for reducing the moisture content from 29% to 13% by using the infrared-hot air drying. The shortest drying time was observed for infrared-hot air drying, followed by infrared and hot air drying. In a study by Afzal et al. (1999), hot air and FIR- hot air was used to dry barley. It was reported that the use of FIR-hot air combination drying of barley required 156%, 238%, and 245% less energy as compared to hot air drying at 40°C, 55°C, and 70°C, respectively.

Utilization of infrared facilitates extraction of many food components (polyphenols, flavonoids, etc.). The potential health benefits of bioactive compounds have led to an increasing interest for extraction studies. Infrared treatment disintegrates the structure and

results in higher extraction yield (Xiang et al., 2022). As compared to convectional extraction methods, infrared assisted extraction leads to enhanced heat transfer efficiency and less energy consumption (Xiang et al., 2022).

In a study by Cheaib et al. (2018), effects of different methods (infrared assisted extraction, microwave assisted extraction, ultrasound assisted extraction, and solid-liquid extraction) on extraction of polyphenols from apricot pomace were investigated. Cheaib et al. (2018) reported that the highest amount of polyphenols (10.8mg gallic acid equivalents/g dry sample) was obtained by using infrared assisted extraction. Increase in polyphenol content by using the infrared assisted extraction was also reported for pomegranate peels and orange peels (El Kantar et al., 2020; Rajha et al., 2019).

Kaveh and Abbaspour-Gilandeh (2020) reported that an increase in infrared power (250, 500 and 750W), air temperature (40, 55 and 70°C), and rotational speed (rates of 5, 10, and 15rpm) resulted in an increase in antioxidant activities of green peas. It has been reported that as the infrared power and air temperature increased, the total phenolic content of the samples increased up to  $12.99 \pm 0.35$  mg GAE g<sup>-1</sup> and the antioxidant capacity increased up to  $75.73 \pm 0.82\%$ .

Irakli et al. (2018) reported that infrared treatment of rice bran at 140 °C for 15 min caused a significant increase in antioxidant activity (DPPH) (from 272.5 mg TE/100g to 385.1 mgTE/100g) and total phenolic content (increased from 7% to 23%).

In a study by Binello et al. (2018), hazelnuts were roasted by using hot air (135°C for 45 min and 195°C for 27 min) or infrared (135°C for 40 min and 195°C for 20 min). Among all samples, hazelnuts infrared roasted at 195°C for 20 min had the highest antioxidant activity (ORAC Assay).

In a study by Sogi et al. (2012), fresh mango samples were infrared treated for 5, 10 or 15 min. It was reported that the samples infrared treated for 5 min (59.23 mg GAE/100 g) and 15 min (71.16 mg GAE/100 g) had higher total phenolic content as compared to the raw (untreated) mango (42.33 mg GAE/100 g). It was reported that the antioxidant activity of the infrared treated samples (338.0 - 416.4 μM TE/100 g fresh weight) was higher as compared to that of control sample (261.5 μM TE/100 g fresh weight).

In a study by Yalcin and Basman (2015), soaked and unsoaked soybean samples were infrared treated at 814 W, 1003 W, 1208 W, 1342 W for 10 min or 15 min. It was reported that higher total phenolic content was obtained for infrared treated samples as compared

to the control sample and the total phenolic content of soybean increased as the infrared power or time increased. The increase in total phenolic content might be due to release of phenolics from the disrupted cell matrix during infrared treatment (Dewanto et al., 2002). It was reported that slight decreases in DPPH radical scavenging activity of soybeans were observed as the infrared power increased.

In a study by Basman (2017), the effects of infrared treatment (735, 1015, 1215, 1310, and 1425 W) (for 6 min) on properties of quinoa samples were investigated. It was reported that quinoa infrared treated at 1215W had significantly higher total phenolic content as compared to other samples (except quinoa infrared treated at 1425W). The phenolic contents of samples infrared treated at 1015W, 1310W, and 1425W were not significantly different than that of control quinoa. The infrared treatment did not cause a significant change in the total flavonoid content of the quinoa samples.

## **2.2. Chia Seed**

The consumption of chia seeds has been increasing over the years. Chia (*Salvia hispanica*) is a pseudocereal with small seed and oval shape. It has a dark brown to beige color with small dark spots on it. Chia seed has high nutritional value and positive impact on human health (Coelho and Salas-Mellado, 2014). Unsaturated fatty acids (omega 3 and omega 6) of chia protect against inflammation, lower cholesterol levels, and enhance cognitive performance (de Falco et al., 2017; Fernandes et al., 2020). Chia seeds are potential source of antioxidants due to phenolic compounds (chlorogenic acid, caffeic acid, myricetin, quercetin, kaempferol, etc.) which are believed to possess cardioprotective and hepatoprotective effects, as well as anti-aging, and anticarcinogenic properties (Ullah et al., 2015). Furthermore, the high amount of dietary fiber in chia seeds is associated with reduced inflammation, lower cholesterol, regulated intestinal function, and promotes weight loss in consumers (Ayerza et al., 2002; de Falco et al., 2017). However, chia seeds must be ingested properly in order to provide the potential health benefits. Since clinical studies about safety and efficiency of chia seeds are limited, safety of the medicinal food or natural food should be validated scientifically (Valdivia-López and Tecante, 2015).

Chia seeds contain approximately 5.80% moisture, 16.54% protein, 30.74% lipids, 34.40% total dietary fiber, and 42.12% carbohydrates (USDA, 2011). Chia seeds contain 631 mg/100 g calcium, 860 mg/100 g phosphorus, 407 mg/g potassium, 16 mg/g sodium,

55.52 µg/100g selenium, 7.72 µg/100g iron, 4.58 µg/100g zinc, and 0.0924 µg/100g copper (Hernández, 2012).

The lipid content of chia seeds was reported as 30.7% (Valdivia-López and Tecante, 2015), 30.74% (Suri et al., 2019), and 40% (de Falco et al., 2017). Chia seeds are rich in omega 3 fatty acids. About 60% of the oil fraction of chia seeds is omega-3 (linolenic acid) while 20% is omega 6 (linoleic acid) (de Falco et al., 2017; Fernandes et al., 2020).

The protein content of chia seeds was reported as 16.5% (Valdivia-López and Tecante, 2015), 18.3% (Coelho and Salas-Mellado, 2014), 18.95% (Parker et al., 2018), 17-20% (Ayerza and Coates, 2005), and 24% (Segura-Campos et al., 2014).

Chia seeds are also rich source of endogenous amino acids such as glutamic acid, aspartic acid, alanine, serine, and glycine. Chia seeds contain ten exogenous amino acids with the highest levels of arginine, leucine, phenylalanine, valine, and lysine. It should be emphasized that chia seeds are gluten-free and can therefore be consumed by celiac patients (Kulczyński et al., 2019).

The total dietary fiber content of chia seeds was reported as 34.4% (Valdivia-López and Tecante, 2015), 39.94-41.41 g/100g seed (dw) (cv. Jalisco) and 36.97-38.79 g/100g seed (dw) (cv. Sinaloa) (Reyes-Caudillo et al., 2008), and 374.4 g/kg seed (Scapin et al., 2016). Chia seeds contain 2.3, 2.6, 8.3, and 9.8 times more fiber per 100 g edible portion as compared to wheat, oat, corn and rice, respectively (Valdivia-López and Tecante, 2015). The ratio of insoluble to soluble dietary fiber is important in terms of nutritional and physiological effects on consumers. American Dietetic Association recommends total dietary fiber intake of 25-30g/day for adults with insoluble/soluble dietary fiber ratio of 3:1 (Borderías et al., 2005). Soluble dietary fiber of chia seed was reported as 2.67 g/100 g (Dhingra et al., 2012), 5-10% (Ali et al., 2012), and 6.16-6.84 g/100g (Reyes-Caudillo et al., 2008). Insoluble dietary fiber of chia seed was reported as  $30.37 \pm 1.02$  g/100 g (Dhingra et al., 2012) and 32.98-34.9 g/100g (Reyes-Caudillo et al., 2008).

Chia seeds contain 50-60 g mucilage /kg of dry seed weight. The mucilage in chia is concentrated in the outer part of the seed coat. Chia mucilage is a water-soluble, complex polysaccharide with high molecular weight and viscosity (Hernández, 2012). Mucilage can be used as a gelling agent, foaming agent, emulsifier, and coating material in food industry due to its functional properties such as water and oil holding capacity and emulsion activity (Reyes-Caudillo et al., 2008).

Chia seeds are rich in phenolic and flavonoid compounds. The total phenolic content of chia seeds was reported as 0.92 mg GAE/g (cv. Jalisco) and 0.88 mg GAE/g (cv. Sinaloa) (Reyes-Caudillo et al., 2008), 0.94 mg GAE/g (Marineli et al., 2014), 97.7 mg GAE/100 g (dw) (Beltrán-Orozco et al., 2020), 1.6398 mg GAE/g (Martínez-Cruz and Paredes-López, 2014), 1.65 mg GAE/g dry sample (Khursheed et al., 2023), 2.14-2.21 mg GAE/g (white chia seeds), and 2.18-2.19 mg GAE/g (dark chia seeds) (Alvites-Misajela et al., 2019). The total flavonoid content of chia seeds was reported as 1.50-1.56 mg CE/g (white chia seeds), 1.54-1.57 mg CE/g (dark chia seeds) (Alvites-Misajela et al., 2019), 0.35 mg QE/100 g dry sample (Khursheed et al., 2023), and 35.8 mg QE/100 g (dw) (Beltrán-Orozco et al., 2020).

It was reported that the high antioxidant activity of chia was due to the presence of phenolic substances such as phenolic acids, isoflavones, and anthocyanins (Martínez-Cruz and Paredes-López, 2014). The radical scavenging activity (% DPPH) of chia seed was reported as 30% (Hatamian et al., 2020), 68.83% (Martínez-Cruz and Paredes-López, 2014), and 88.27% (Ghafoor et al., 2020). Chlorogenic acid and caffeic acid are known to inhibit lipid peroxidation. As compared to vitamin E and vitamin C, caffeic acid and chlorogenic acid exhibit higher antioxidant activity. This makes chia a well-balanced and reliable source of omega-3 fatty acids (Valdivia-López and Tecante, 2015).

### **2.2.1. Functional Properties of Chia**

The most significant functional properties of proteins in foods are; water and oil holding capacity, foaming capacity, and stability, emulsion activity and stability. High amounts of protein and fiber in chia results in high water and oil holding capacity, as well as emulsion activity (Olivos-Lugo et al., 2010).

In literature, water holding capacity of chia seed was reported as 5 g water/g sample (for whole chia seed), 5.5 g water/g sample (for chia seed powder) (Darwish et al., 2018), 5.2 g water/g sample (Haripriya, 2020), 6g/100g (Hatamian et al., 2020), 10.64 g/g (solvent meal), and 10.58 g/g (pressed meal) (Capitani et al., 2012). Water soluble and insoluble fiber of chia has the ability to retain water. Chia mucilage is a water-soluble, complex polysaccharide with high molecular weight and viscosity (Hernández, 2012). Alfredo et al. (2009) reported that high lignin and hemicellulose content in chia seeds have a positive effect on water holding capacity.



Oil holding capacity is an important property in development of new food products with long shelf life (Kinsella and Melachouris, 1976). Oil holding capacity of chia seed was reported as 2.03 g/g (solvent meal) and 1.26 g/g (pressed meal) (Capitani et al., 2012), 3.9 g/100g (Hatamian et al., 2020), 2.23 ml/g (García-Salcedo et al., 2018), 2.7 ml/g (Haripriya, 2020), and 3.25 g oil/g sample (for whole chia seed), 2.5 g oil/g sample (for chia seed powder) (Darwish et al., 2018).

Proteins are generally known to possess strong emulsifying properties and the high protein content of chia seeds may enhance their emulsifying activity (Haripriya, 2020). In literature, emulsion activity of chia was reported as 14.69% (García-Salcedo et al., 2018) and 42% (Haripriya, 2020).

### **2.3. Chia Mucilage**

Mucilage is generally confused with gums but these two terms are different. Gums are pathological products usually formed by plants after injury (e.g. drought) or damage to the cell wall, while mucilages are physiological products of metabolism that are normally produced (Jani et al., 2009).

Mucilage obtained from plant seeds and other parts of the plant is complex polysaccharides consisting of monosaccharides and uronic acid units. Nowadays, utilization of plant seed mucilages in the food industry is increasing because of the advantages of synergistic effects of commercial hydrocolloids (Vardar et al., 2021). They are used as thickening, emulsifying, and gelling agent in industry.

Although the chemical structure of chia seed mucilage has not been fully explained, Lin et al. (1994) proposed a tentative structural model suggesting that the polysaccharide is composed of a tetrasaccharide structure. In this model, 4-O-methyl- $\alpha$ -D-glucopyranosyl residues occurring as branch at O-2 of some  $\beta$ -D-xylopyranosyl residues in main chain, which consists of (1 $\rightarrow$ 4) - $\beta$ -D-xylopyranosyl- (1 $\rightarrow$ 4)- $\alpha$ -D-glucopyranosyl- (1 $\rightarrow$ 4)- $\beta$ -D-xylopyranosyl units.

The mucilage in chia is concentrated in the outer part of the seed coat. Chia seeds contain 50-60 g mucilage/kg of dry seed weight (Hernández, 2012). Composition of chia mucilage was reported as follows; moisture content as 11.3% (Cuomo et al., 2020), 115g/kg (dw) (Capitani et al., 2013) and %15.15 (Hernández, 2012), protein content as %9.7 (Cuomo et al., 2020), 10.63 g/100g (dw) (Fernandes and Salas-Mellado, 2017), 112 g/kg (dw) (Capitani et al., 2013), fat content as 0.55% (dw) (Coorey et al., 2014), 1.78%

(Hernández, 2012), 2.05 g/100g (dw) (Fernandes and Salas-Mellado, 2017), 31 g/kg (dw) (Capitani et al., 2013), ash content as 6.8% (Cuomo et al., 2020), 8.07% (Hernández, 2012), 85g/kg (dw) (Capitani et al., 2013), and total dietary fiber content as 57.84% (dw) (Coorey et al., 2014) and 75.3% (Cuomo et al., 2020). Hernández (2012) reported polysaccharide content in chia seed mucilage as 71.22%. In literature, polysaccharide content of guar gum was reported as 23.7% and xanthan gum as 98%.

### **2.3.1. Chia Mucilage Extraction**

The basic procedure for mucilage extraction from chia seeds involves treating the seeds with a specific amount of water. After extraction, aqueous suspension is dried and ground to a certain particle size. Important parameters for mucilage extraction include seed:water ratio, extraction temperature-time and pH (Hernández, 2012). In literature, the effects of conventional heat treatment, sonication and microwave on mucilage extraction were studied.

In a study by Hernández (2012), mucilage extraction from chia seeds was carried out for 2 hours at different seed:water ratios (1:20, 1:30, and 1:40), pH values (4, 6, and 8) and temperatures (4, 40, and 80°C). Increasing the temperature from 20°C to 80°C increased the mucilage yield, irrespective of the seed:water ratio. When the pH was increased from 4 to 8, mucilage extraction yield (%) increased. The mucilage samples were dried at 50°C and highest yield (6.97%) was reported for seed:water ratio of 1:40, at 80°C and pH 8. The mucilage has 15% moisture, 48% carbohydrate, 23.22% uronic acid, 8% ash, 4% protein, and 1.78% lipid.

In a study by Tavares et al. (2018), hot and cold extraction methods were used for mucilage extraction. Extraction method reported by Hernández (2012) for hot extraction, which involved a seed:water ratio of 1:40, pH 8 and temperature of 80°C for 2 hours was used. The mucilage samples were dried at 50°C. For cold extraction method, various seed:water ratios (1:10, 1:20, 1:30, 1:40), pH 8, 27°C for 2 hours was used. Controlled pressure of 313 kgf/cm<sup>2</sup> was used to separate the mucilage from the seeds and the samples were freeze-dried. The mucilage yield for hot extraction was reported as 6.52%. For cold extraction method, higher mucilage yields were obtained as 8.46%, 8.65%, 8.31% for 1:20, 1:30, and 1:40 seed:water ratios, respectively. It has been stated that the reason for higher mucilage yield as compared to other methods used in the literature may be the

better separation of seeds and mucilage by using controlled pressure and the easy removal of dry mucilage without sticking to the surface of the container as a result of freeze drying. Castejón et al. (2017) extracted the mucilage from chia seeds at a seed:water ratio of 1:40, with constant mixing at 50°C for 2 hours and sonication with a power of 30% for 3 min. The yield of chia mucilage was found to be 6.52%.

In a study by Muñoz-Tebar et al. (2021), the extraction was carried out at a seed:water ratio of 1:40, 80°C, and pH 8. The mucilage yield was 6.6% for oil-free chia seeds and 15.3% for defatted chia flour.

Nayani (2020) used microwave assisted extraction method in order to obtain mucilage from chia seeds. Mucilage was used as a fat replacer in biscuits. The mucilage was extracted by keeping the chia seeds in water (80°C) in the microwave at 1.3 W/gm for 15 minutes. The mucilage yield obtained by the microwave-assisted extraction method was reported as 8%. The extraction time was 4 times shorter than the time used in the study of Muñoz et al. (2012).

Lamia et al. (2016) extracted mucilage from Indian fig (*Opuntia ficus-indica*) by using microwave power in the range of 100-1000W. The mucilage yields were reported as 8.03%, 8.81%, and 8.95% for 500W-7 min, 700W-5 min, and 900W-3 min samples, respectively.

Amutha Gnana Arasi et al. (2016) extracted mucilage from apple guava fruit (*Psidium guajava*) by using microwave-assisted extraction method. The aqueous suspension of ground fruit was heated by using microwave power in the range of 140-200 W. There was a linear increase in mucilage yield with the increase in microwave power from 140W to 200W. The increase in microwave power improved the solubility of the sample resulting in increased extraction efficiency.

In a study by Shah and Seth (2011), okra (*Abelmoschus Esculentus*) was soaked in water for 24 hours and then microwave at 160W-800W for 3-60 min was applied for extraction. The amount of mucilage obtained from 5 g of sample after 1 hour of extraction by using convectional method was 0.476 g. As compared to convectional extraction method, the yield (0.531 g for 5 g sample) of mucilage was found to be higher in microwave extraction at 160W-40 min. The microwave extraction applied at 160W for 40 min resulted in an 11.55% increase in mucilage yield in a shorter time as compared to the 1 hour conventional extraction.

### **2.3.2. Functional Properties of Chia Mucilage**

Hydrocolloids are widely used in the food industry due to water holding capacity. Gum and mucilage are subgroup of hydrocolloids (Tosif et al., 2021). Water holding capacity of gums and mucilages can be attributed to the hydroxyl groups in their structure, as well as the presence of protein substituents (Alpizar-Reyes et al., 2017). Chia seeds contain 5-6% mucilage. Chia mucilage is a water-soluble, complex polysaccharide with high molecular weight and viscosity (Hernández, 2012).

Oil plays an important role in baked products by providing dough greasing, air incorporation and preventing gas-bubble coalescence during mixing (Felisberto et al., 2015; Fernandes and Salas-Mellado, 2017). Oil holding capacity is the most important functional property of hydrocolloids. The high oil holding capacity of mucilages extracted from fruits or seeds is attributed to the presence of monopolar molecules. Chia mucilage can be used instead of fat in food formulations due to its moisture retention capability and its effect on viscosity increase. Mucilages with high oil holding capacity could also enhance the texture of foods (Tosif et al., 2021).

Mucilage and gums are notable for their use in the pharmaceutical and food industries due to their excellent emulsifying properties. They are used in these sectors for the suspension of particles, stabilization of emulsions, control of crystallization, encapsulation, thickening, and film formation (Tosif et al., 2021). The lipid content of chia mucilage can play a significant role in stabilizing oil-water emulsions.

Chia mucilage is used as a substitute in bakery products, cereals, dairy products, and meat products. Fernandes et al. (2021) used chia mucilage as a substitute for fat (margarine) in the commercial cake formulation and 43.5% reduction in the final lipid content of the cake was obtained. Chia mucilage can be used instead of emulsifiers and stabilizers in ice cream while maintaining the quality of the product (Campos et al., 2016). In a study by Punia and Dhull (2019), chia mucilage was added to cookie formulations at ratios of 10, 20, 30, and 40%. The addition of chia mucilage to cookie formulation instead of oil did not affect the technical properties (rheology) of the cookies and the addition of mucilage reduced the caloric value (Punia and Dhull, 2019).

Fernandes and Salas-Mellado (2018) reported that chia mucilage can be used as a substitute for oil or egg yolk in mayonnaise. Substitution of egg yolk was found to be more acceptable than substitution of oil in the formulation.

### 3. MATERIALS AND METHODS

#### 3.1. Materials

Chia seeds (*Salvia hispanica*) used in the study were purchased from “Yayla Legumes” (Ankara, Turkey). Quercetin (Sigma PHR1488), caffeic acid (Sigma C0625), chlorogenic acid (Cayman Chemicals-70930), Trolox (Sigma 238813), rutin hydrate (Sigma R5143), ferulic acid (Sigma PHR1791), p-Coumaric acid (C9008) were used as standards in the study.

In the present study, the following chemicals were used; sodium hydroxide (Sigma 30620), sulfuric acid (ISOLAB 970.026), ethanol (ISOLAB 920.026), methanol (Sigma 34885), hydrochloric acid (ISOLAB 932.106), aluminum chloride (Sigma 237051), ammonium acetate (Sigma 238074), acetic acid (Supelco 100063), acetone (ISOLAB 901.027), acetonitrile (Sigma 34851), copper (II) dichloride dihydrate (Sigma 307483), copper (II) sulfate (911.026), boric acid (Sigma 100165), Folin&Ciocalteu's phenol reagent (Sigma F9252), gallic acid (Sigma G7384), neocuproine (Sigma N1501), sodium carbonate (Sigma 106392), sodium nitrite (Sigma S2252) and sodium acetate (Sigma 79714).

#### 3.2. Infrared Treatment

In the present study, a laboratory scale infrared equipment with a closed drying chamber fitted with twelve 150W halogen lamps (Infrared, BR125 IR; Philips, Eindhoven, The Netherlands) and two aeration channels (12V each) was used. The system has ventilation channels to prevent heating and provide air circulation. The wavelength spectrum of halogen lamps was 0.2–4 $\mu$ m, with a pronounced peak at approximately 1 $\mu$ m. Aluminum reflectors were used on the walls of the equipment to prevent light absorption. The distance between the light source and the sample could be adjusted between 5 cm and 39 cm. The ambient temperature inside the infrared equipment was measured with a thermocouple.

In preliminary studies, chia seeds were infrared treated at 600W, 800W, and 1000W for 30 and 60 minute. As the infrared power increased, an increase in total phenolic content was observed. Therefore, infrared power of 1200W was also applied to chia seeds. It was observed that infrared treatment at 1200W for 60 min and 1100W for 60 min resulted in burnt chia seeds. According to the preliminary results, it was decided to apply infrared

treatment at 700W, 900W, and 1100W for 25 and 50 minutes in the study. The distance between the tray and light source was set to 20 cm in the present study.

Infrared treatment at each power and time was carried out as seven replicates. For each replication, 140 g chia samples were spread on a greaseproof paper placed on a 39x40 cm wooden tray with sticks on each side in order to obtain a homogeneous thickness of 2-3 mm for chia samples (Figure 2). During the infrared treatment at each power and time, the surface temperatures of the samples were measured in every 5 min at 4 different points by an IR thermometer (Raytek MX6, Germany). The average surface temperatures of the chia samples during the infrared treatment and internal temperature in the infrared equipment at the beginning and at the end of each treatment is given in Table 1. The average surface temperatures of the chia samples infrared treated at 700W-25 min, 700W-50 min, 900W-25 min, 900W-50 min, 1100W-25 min, and 1100W-50 min were 113.5°C, 119.4°C, 139.6°C, 148.3°C, 160.8°C, and 168.6 °C, respectively (Table 1).



**Figure 2.** Chia seeds prepared for infrared treatment

Seven replicates for each infrared power-time treatment were combined in a single large batch and then ground using a Multi-Purpose Disintegrator (IC-04A, China). The ground chia was then sifted through a 850µm sieve. The sample above the 850µm sieve was ground again by using Multi-Purpose Disintegrator (IC-04A, China) and sifted again through 850µm sieve. Since the amount of sample over 850 µm sieve is less, a blender (Arzum Bebbe, AR 854) was used for further grinding. The ground chia sample (<850µm) was stored at +4°C until analysis.

**Table 1.** The surface and internal temperatures during infrared treatment

Infrared Power - Time	Internal Temperature (°C) (Beginning-end)	Average surface temperature* during process (°C)
700 W-25 min	58-63	113.5
700 W-50 min	58-65	119.4
900 W-25 min	72-77	139.6
900 W-50 min	72-80	148.3
1100 W-25 min	84-91	160.8
1100 W-50 min	84-95	168.6

\*Average surface temperatures are given as the average of means for surface temperatures of seven infrared treatment replicates. (For each infrared treatment, means for surface temperature was calculated as the average of measurements taken in every 5 min at 4 different points.)

### 3.3. Analyses of Chia Samples

#### 3.3.1. Moisture Content

Moisture contents of control and infrared treated chia samples (<850 $\mu$ ) were determined at 135°C by using AACC Method No 44-19 (Air oven method at 135°C) (AACC, 1990). The results were given as the average of two values.

#### 3.3.2. Ash Content

Ash content of control and infrared treated chia samples (<850 $\mu$ ) were determined according to AACC Method No 08-01 (AACC, 1990). The results were given as the average of two values.

#### 3.3.3. Protein Content

Protein content (Nx6.25) of control and infrared-treated chia samples (<850 $\mu$ ) were determined by using Kjeldahl method (AACC Method 46-12) (AACC, 1990).

Sulfuric acid (25ml) and catalyst (3-5 g Cu<sub>2</sub>SO<sub>4</sub>: K<sub>2</sub>SO<sub>4</sub>, (7:3 w/w ratio)) was added to 1g chia sample in Kjeldahl digestion tubes. After preheating at 250°C for 45 minutes, combustion is performed at 425°C until green color formation (~1-1.5 hours). NaOH solution (33%, w/v) and boric acid solution (4%, w/v) were used for distillation. Tashiri indicator was added and the samples were titrated by using 0.1 N HCl. Protein content (% dw) was calculated by using the equation given below. Conversion factor was taken as 6.25. The results were given on dry basis as the average of two values.

Total nitrogen content (%N) =  $((0.014 \times N \times V) \times 100) / W$

Protein content (% , dw) = Total nitrogen content (%N) x F

- N = Normality of HCL solution (0.1N)
- V = Volume of HCl used for titration of sample (ml)
- W = Sample weight (g)
- 0.014 = Milliequivalent weight of nitrogen
- F = Conversion factor (6.25)

### 3.3.4. Color Analysis

Color values of control and infrared-treated chia seeds were analyzed with Minolta Spectrophotometer (CM-3600d). The color values were evaluated in terms of CIE L\*(lightness/darkness); a\*(redness/greenness) and b\*(yellowness/blueness). The results were given as the average of three values.

### 3.3.5. Total Dietary Fiber Analysis

The total dietary fiber content was determined using Megazyme Total Dietary Fiber Kit (Megazyme International Ireland Ltd., Ireland), by using modified method of the AOAC 991.43, AOAC 985.29, AACC 32-07, and AACC 32-05 methods.

Since the fat content of chia is greater than 10%, fat was removed prior to analysis as indicated in the test procedure. For defatting, the ground chia (<850 $\mu$ m) was mixed with hexane with a ratio of 1:5 (w/v) and stirred with a magnetic stirrer for 1 hour to remove the oily portion, as described by Rahman et al. (2017). After removal of hexane at room temperature, moisture content was determined. The defatted chia sample (0.25g) was used in the analysis.

The aim of the procedure is to remove starch and protein from sample by using  $\alpha$ -amylase, protease, and amyloglucosidase enzymes. At the first step starch was hydrolyzed with  $\alpha$ -amylase at 98-100 $^{\circ}$ C and then protein was hydrolyzed by protease at 60 $^{\circ}$ C and finally starch was broken down into glucose units at 60 $^{\circ}$ C with amyloglucosidase. To precipitate soluble fibers and remove protein and glucose units, ethanol (95%) was added to the samples and filtration was carried out. The filtrate was washed with ethanol (78% and 95%) and acetone and then dried overnight at 105 $^{\circ}$ C.

Ash analysis was performed on the dried samples at 525 $^{\circ}$ C. The amount of ash was subtracted from the residue obtained after drying overnight at 105 $^{\circ}$ C to ensure that the



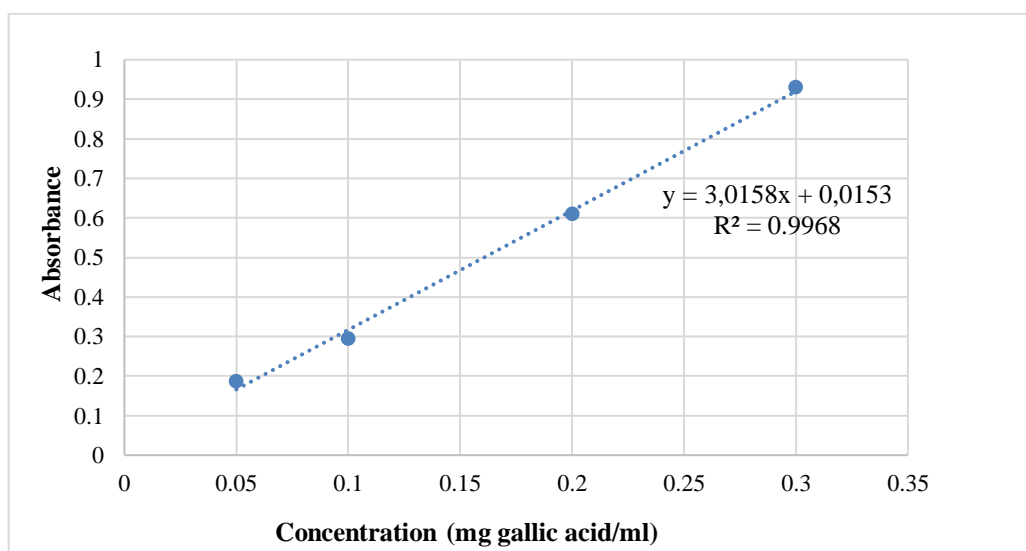
amount of ash did not affect the total amount of dietary fiber. The results were given on dry basis as the average of two values.

### 3.3.6. Total Phenolic Content

Total phenolic content of chia samples was determined using the Folin-Ciocalteu method with some modifications described by (Beltrán-Orozco et al., 2020).

Chia sample extract was prepared by using Beltrán-Orozco et al. (2020) method with some modifications. Chia sample (1g, dw) was mixed with 10ml of 80% ethanol at room temperature for 4 hours. The samples were centrifuged at 6000g at 4°C for 15 minutes. The extract was filtered through Whatman No.1 filter paper and total phenolic content of the extract was determined immediately.

To determine the total phenolic content of chia samples, 0.06ml of chia extract was mixed with 1.54ml of distilled water. Then, 100µl of Folin Ciocalteu reagent was added, vortexed for 10 seconds and left at room temperature for 3 minutes. Na<sub>2</sub>CO<sub>3</sub> solution (300µL, 10% w/v) was added, vortexed for 10 seconds and rested at room temperature for 60 minutes in a dark place. The absorbance was measured at 765 nm. The calibration curve for the gallic acid standard (0-300ppm (mg/L)) is given in Figure 3. The results were given as the average of four values and expressed as mg GAE/g dry sample.

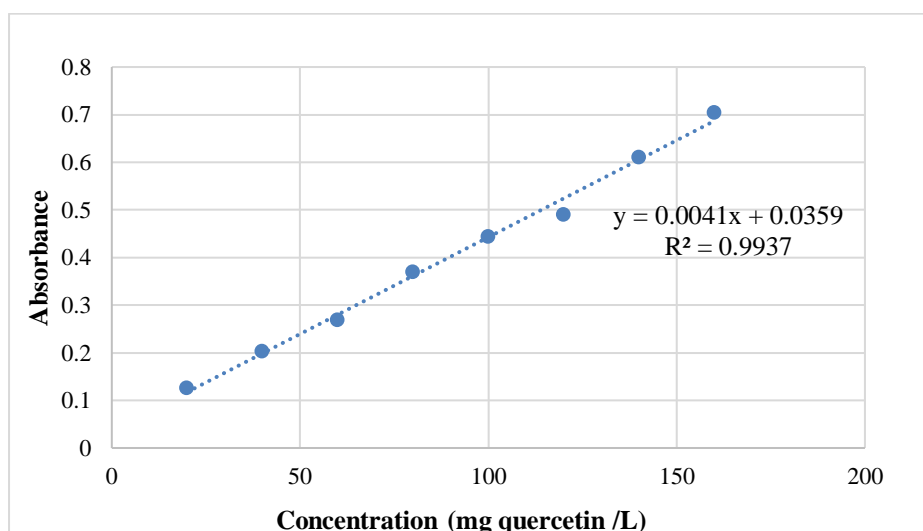


**Figure 3.** Calibration curve of gallic acid standard

### 3.3.7. Total Flavonoid Content

Chia sample extract used in total phenolic content analysis was also used for total flavonoid content determination.

Total flavonoid content was determined using the method described by Scapin et al. (2016). Initially, 250 $\mu$ l of chia extract was mixed with 1250 $\mu$ l of distilled water. Then, 75 $\mu$ l sodium nitrite (NaNO<sub>2</sub>) solution (5%) was added and the mixture was allowed to stand for 5 minutes. Then, 150 $\mu$ l aluminum chloride (AlCl<sub>3</sub>) solution (10%) was added and the mixture was left to stand for 5 minutes. 500 $\mu$ l sodium hydroxide (NaOH) (1M) and 775 $\mu$ l distilled water was added and mixed thoroughly. The absorbance was measured immediately at 510 nm. The calibration curve for the quercetin standard (0-160ppm) is given in Figure 4. The results were given as the average of three values and expressed as mg quercetin/g dry sample.



**Figure 4.** Calibration curve of quercetin standard

### 3.3.8. Antioxidant Activity

#### 3.3.8.1. DPPH Assay

Chia sample extract used in total phenolic content analysis was also used in DPPH assay. Radical scavenging activity (%) was determined by using the method described by Laczkowski et al. (2018).

DPPH (2,2-Diphenyl-1-picrylhydrazyl) solution was prepared with 80% ethanol at a concentration of 6x10<sup>-5</sup>M. 3.9ml DPPH solution (6x10<sup>-5</sup>M) was added to 0.1ml of chia extract, the mixture was vortexed and kept in a dark place for 30 minutes.

For the preparation of the control sample, 3.9ml DPPH solution was added to 0.1 ml ethanol (80%), instead of chia extract. The absorbance of chia samples and control was measured at 515nm against 80% ethanol (blank). The radical scavenging activity (%) was calculated according to the equation given below and the results were given as the average of three values.

$$\text{Radical scavenging activity (\%)} = \left(1 - \frac{\text{absorbance of sample}}{\text{absorbance of control}}\right) \times 100$$

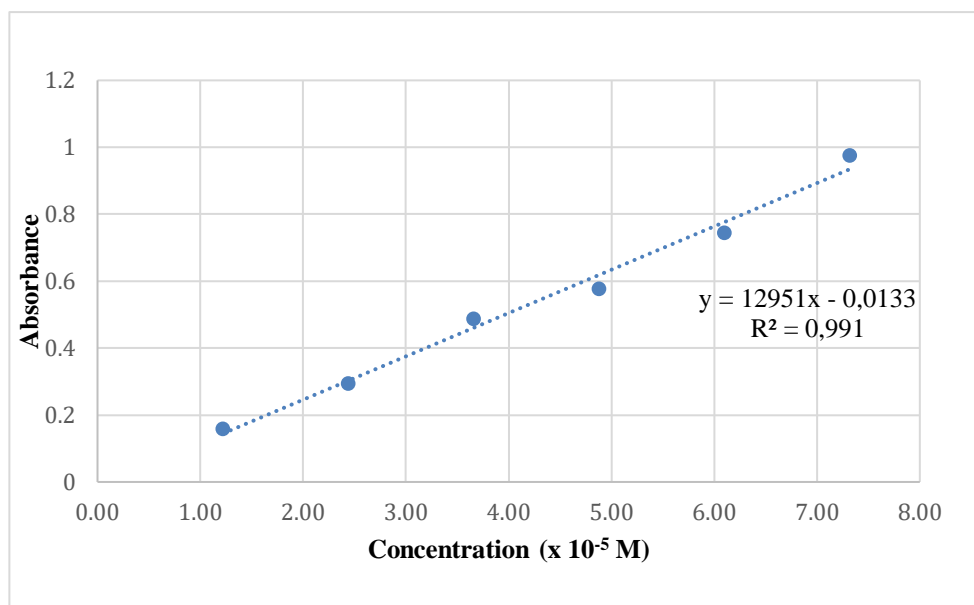
### 3.3.8.2. QUENCHER-CUPRAC Method

QUENCHER-CUPRAC method described by Tufan et al. (2012) was used with some modifications in order to determine the total antioxidant capacity of chia samples. 100mg (dw) chia sample (<850 $\mu$ ) was first mixed with 900mg (dw) cellulose in order to obtain a ratio of 1/10 (w/w). Chia sample (10mg) was taken from this mixture and mixed with 4.1ml of CUPRAC reagent solution. CUPRAC reagent solution was prepared by adding copper (II) solution (1ml), neocuproin solution (1ml), ammonium acetate solution (1ml) and water:ethanol (1.1 ml, 1:1 v/v). After vortexing for 1 minute, the mixture was shaken at room temperature for 30 minutes. Sample was centrifuged at 8000rpm for 2 minutes and filtered through a 0.45 $\mu$ m micro-filter (Chromafil GF/PET-45/25). Absorbance was measured at 450nm. The calibration curve for Trolox standard (1.22x10<sup>-5</sup>–7.32x10<sup>-5</sup> M) is given in Figure 5.

Trolox stock solution was prepared at a concentration of 1.0x10<sup>-3</sup>M, and standard solutions in the range of 1.22x10<sup>-5</sup>–7.32x10<sup>-5</sup>M were prepared from this stock solution. Standard curve was obtained by using the mixtures including Trolox solution (x ml), 1ml CuCl<sub>2</sub>, 1ml neocuproin, 1ml ammonium acetate and ethanol:water (1.1- x ml). For blank, 1ml of CuCl<sub>2</sub>, 1ml of neocuproin, 1ml of ammonium acetate and 1.1ml of ethanol:water was mixed. Absorbance was measured at 450nm. Trolox standard curve was plotted (Figure 5) and coefficient of x in the equation was used as  $\epsilon_{TR}$  (molar absorption coefficient of Trolox). Total antioxidant capacity (TAC) was calculated in mmol Trolox Equivalent (TE)/kg sample using the equation given below. The results were given as the average of two values.

$$\text{Total antioxidant capacity (TAC, mmol/kg)} = \frac{\text{Absorbans}}{\epsilon_{TR}} \times \frac{\text{Total volume (ml)}}{\text{m (sample)(kg)}} \times \text{DF} \times 10^3$$

- A : Sample absorbance measured at 450nm
- $\epsilon_{TR}$  : Molar absorption coefficient of Trolox in CUPRAC method (Coefficient of x in the equation given in calibration curve for Trolox standard) (12951 L/mol.cm)
- Total volume : Total volume of CUPRAC solution (4.1ml)
- DF : Dilution factor (10)
- m : Amount of sample taken (kg)



**Figure 5.** Calibration curve of trolox standard

### 3.3.9. Phenolic Profile of Chia Samples

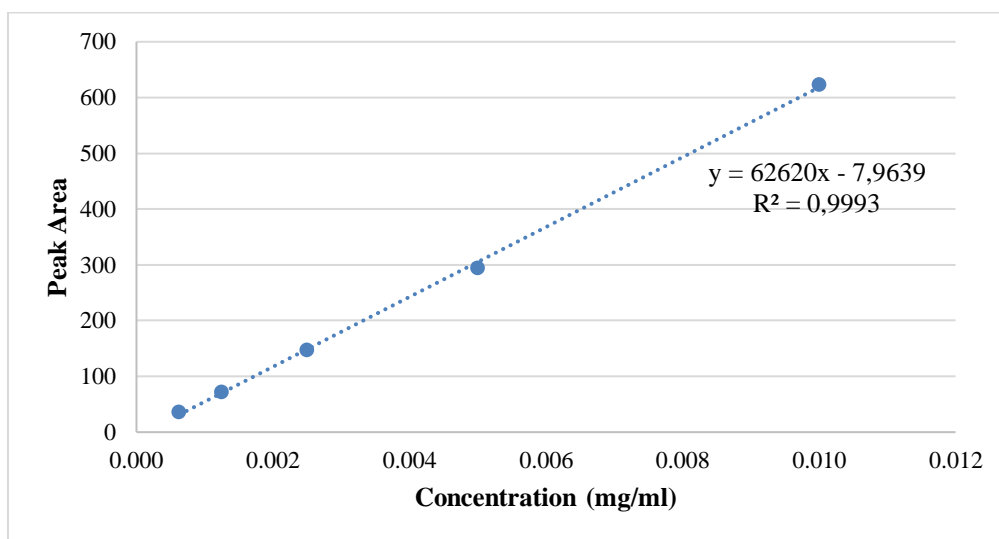
**Extraction:** Chia sample (1g, dw) was mixed with 80% methanol for 6 hours. In preliminary studies, extraction for 2, 4, 6, and 8 hours was carried out and the highest phenolic composition was obtained for the extraction time of 6 hour. The sample was centrifuged at 4°C, 6000g for 15 minutes. The extract was filtered through Whatman No.1 filter paper and analyzed immediately (Abdel-Aty et al., 2021).

**HPLC analysis:** Caffeic acid, chlorogenic acid, rutin, p-Coumaric acid, ferulic acid, and quercetin of chia samples was determined by using modified method of Pająk et al. (2019) HPLC (Agilent Technologies, Waldbronn, Germany) equipped with a Multiple Wavelength Detector (MWD) (Agilent Technologies, Waldbronn, Germany) was used.

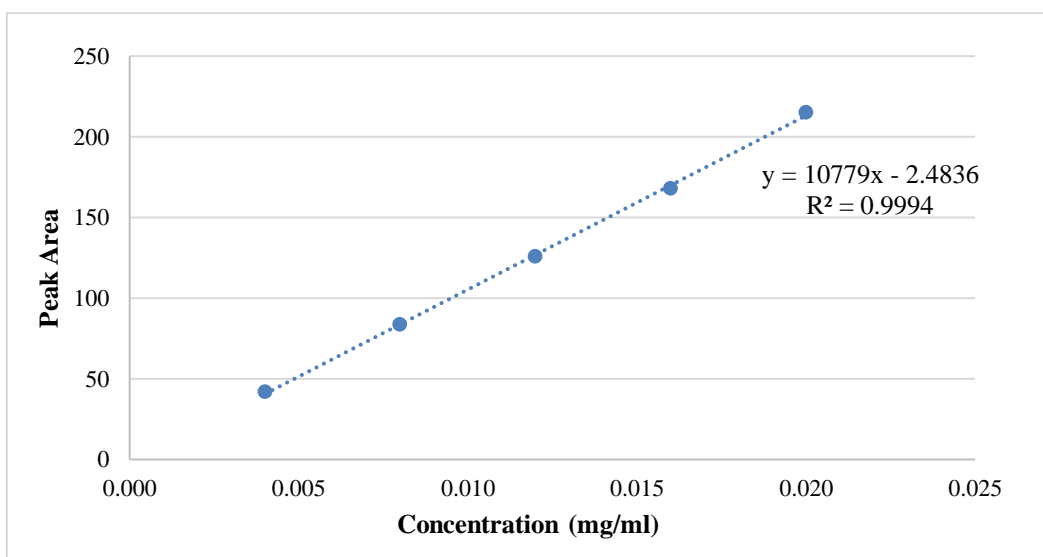
The sample was analyzed on C18 column (250mm x 4.6mm x 5µm, Supelco, Germany) at a temperature of 30°C. Chromatographic separation was performed at a flow rate of

1.0 ml/min with gradient elution by using two solvents for mobil phase: solvent A- 2% acetic acid and solvent B-acetonitrile. The gradient elution used was as follows; 97% B at 0 min, 92% B at 10 min, 85% B at 20 min, 80% B at 30min, 70% B at 40min, and 60% B at 50min. Injection volume was 10  $\mu$ L. Chlorogenic acid was detected at  $\lambda$ =280 nm, while other phenolics (caffeic acid, p-Coumaric acid, rutin, ferulic acid, and quercetin) was detected at  $\lambda$ =320 nm. The results were given as the average of four values.

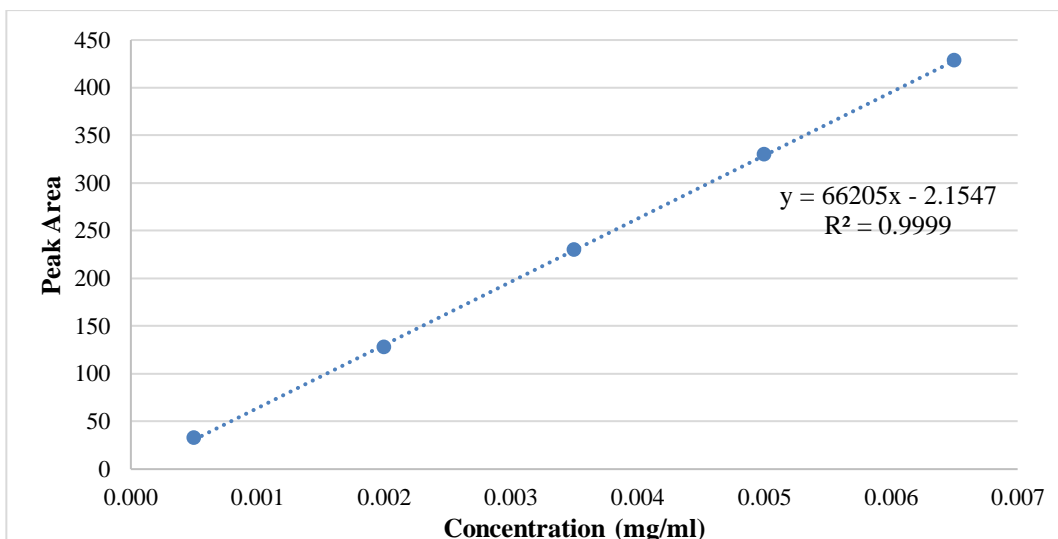
Standard for caffeic acid (0.00625- 0.01 mg/ml), chlorogenic acid (0.002- 0.02 mg/ml), rutin (0.002-0.042 mg/ml), p-Coumaric acid (0.0002-0.0042 mg/ml), ferulic acid (0.00025 -0.0055 mg/ml), and quercetin (0.005-0.03 mg/ml) was prepared by using methanol (80%). Calibration curves of the standards are given in Figure 6-11.



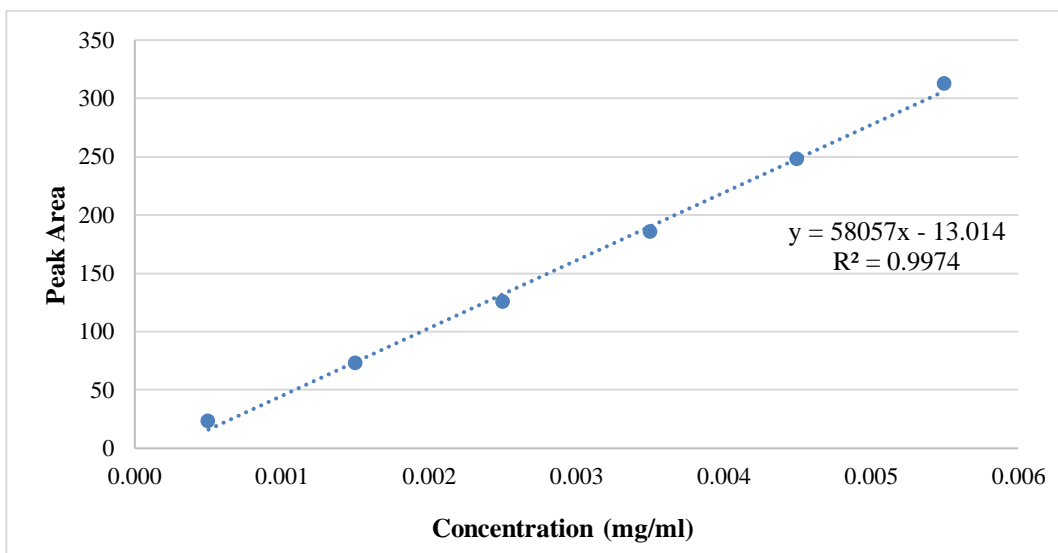
**Figure 6.** Calibration curve of caffeic acid standard



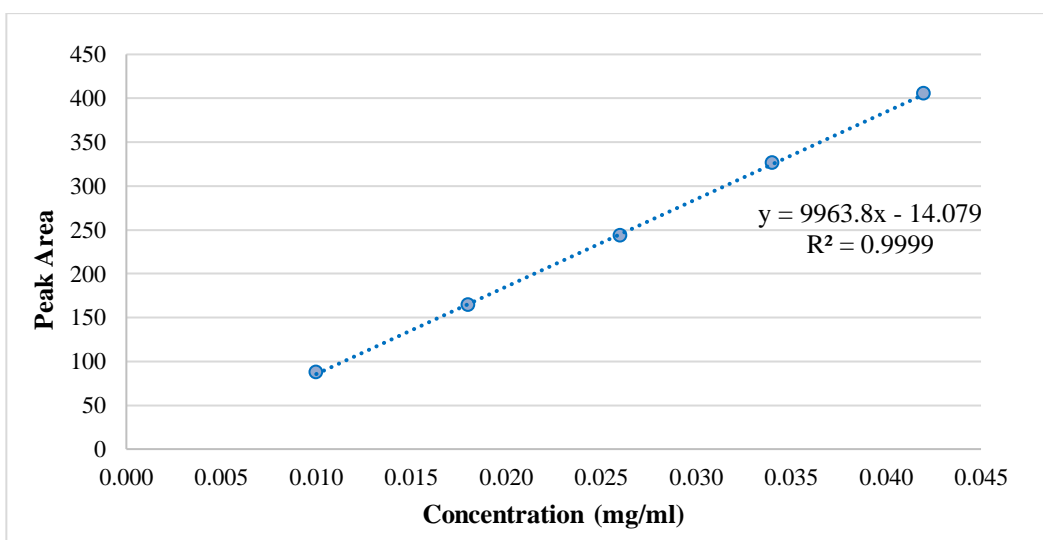
**Figure 7.** Calibration curve of chlorogenic acid standard



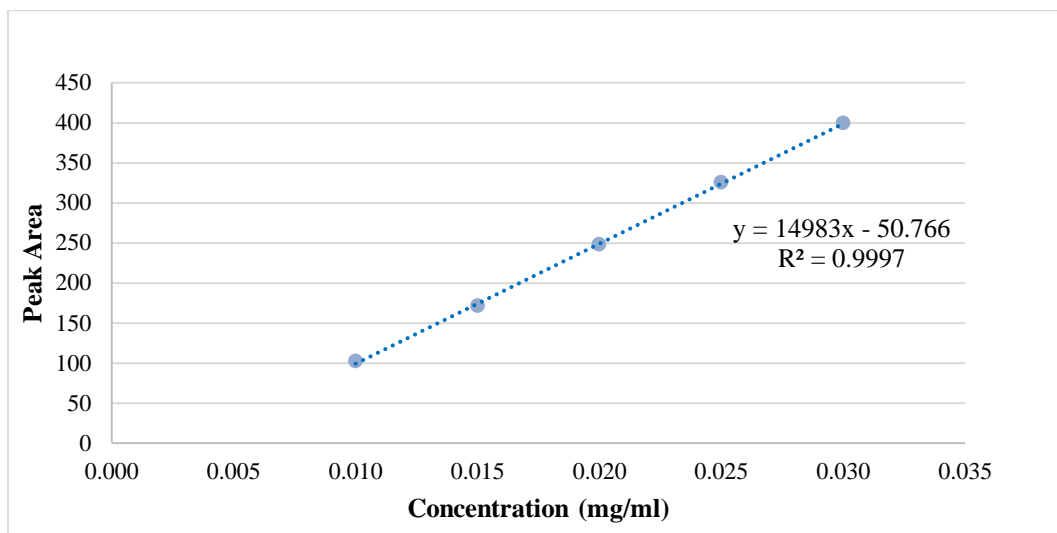
**Figure 8.** Calibration curve of p-Coumaric acid standard



**Figure 9.** Calibration curve of ferulic acid standard



**Figure 10.** Calibration curve of rutin standard



**Figure 11.** Calibration curve of quercetin standard

### **3.3.10. Functional Properties of Chia Samples**

#### **3.3.10.1. Water Holding Capacity**

Water holding capacity of control and infrared treated chia samples was determined by using the method of Alfredo et al. (2009). 10ml of distilled water was added to chia sample (1g, dw) while vortexing (for 1 minute) at room temperature and then centrifuged at 2200g for 30 minutes. The water holding capacity was expressed as the grams of retained water per gram of dry sample. The results were given as the average of three values.

#### **3.3.10.2. Oil Holding Capacity**

Oil holding capacity of control and infrared treated chia samples was determined by using the method of Alfredo et al. (2009). Chia sample (1g, dw) was mixed with 10ml corn oil for 1 minute at room temperature and then centrifuged at 2200g for 30 minutes. The density of the corn oil used in the study was 0.9056 g/ml. The oil holding capacity was expressed as the grams of retained oil per gram of dry sample. The results were given as the average of three values.

#### **3.3.10.3. Emulsion Activity and Emulsion Stability**

Emulsion activity and emulsion stability of control or infrared treated chia samples was determined by using the method of García-Salcedo et al. (2018), with some modifications.

In order to determine the emulsion activity, chia sample (1g, dw) was mixed with 20ml distilled water and homogenized (IKA Ultraturrex, Germany) at 10000rpm for 25 seconds. Then, 7ml corn oil was added and homogenized again for 25 seconds. The density of the corn oil used in the study was 0.9056 g/ml. After centrifugation at 4000rpm for 9 minutes, the layers of liquid, emulsion, and oily phase was detected over the sample precipitate. The height of the emulsion layer was measured. The emulsion activity (%) was calculated by using the equation given below. The results were given as the average of three values.

$$\text{Emulsion activity (\%)} = \frac{\text{Height of the emulsified layer}}{\text{Height of the tube}} \times 100$$

The tubes for which the emulsion activity calculated were heated at 80°C for 30 minutes, then cooled to room temperature and centrifuged at 4000rpm for 10 minutes. The height of the emulsion layer was measured. The emulsion stability (%) was calculated by the equation given below (Yalcin, 2011). The results were given as the average of three values.

$$\text{Emulsion stability (\%)} = \frac{\text{Height of the emulsified layer}}{\text{Height of the tube}} \times 100$$

### **3.3.11. FTIR Spectroscopy**

The control and infrared treated chia samples were characterized by using Attenuated Total Reflectance-Fourier Transform Infrared (ATR-FTIR) (Thermo Scientific, Nicolet™ iS50, Waltham, MA) at HUNITEK (Hacettepe Üniversitesi İleri Teknolojiler Uygulama ve Araştırma Merkezi, Ankara, Turkey). Analysis was carried out as 16 scanning frequencies over a wavenumber range of 4000-400 cm<sup>-1</sup> with 4 cm<sup>-1</sup> resolution. This technique provided information about the different chemical groups present in the chia samples.

### **3.4. Mucilage Extraction**

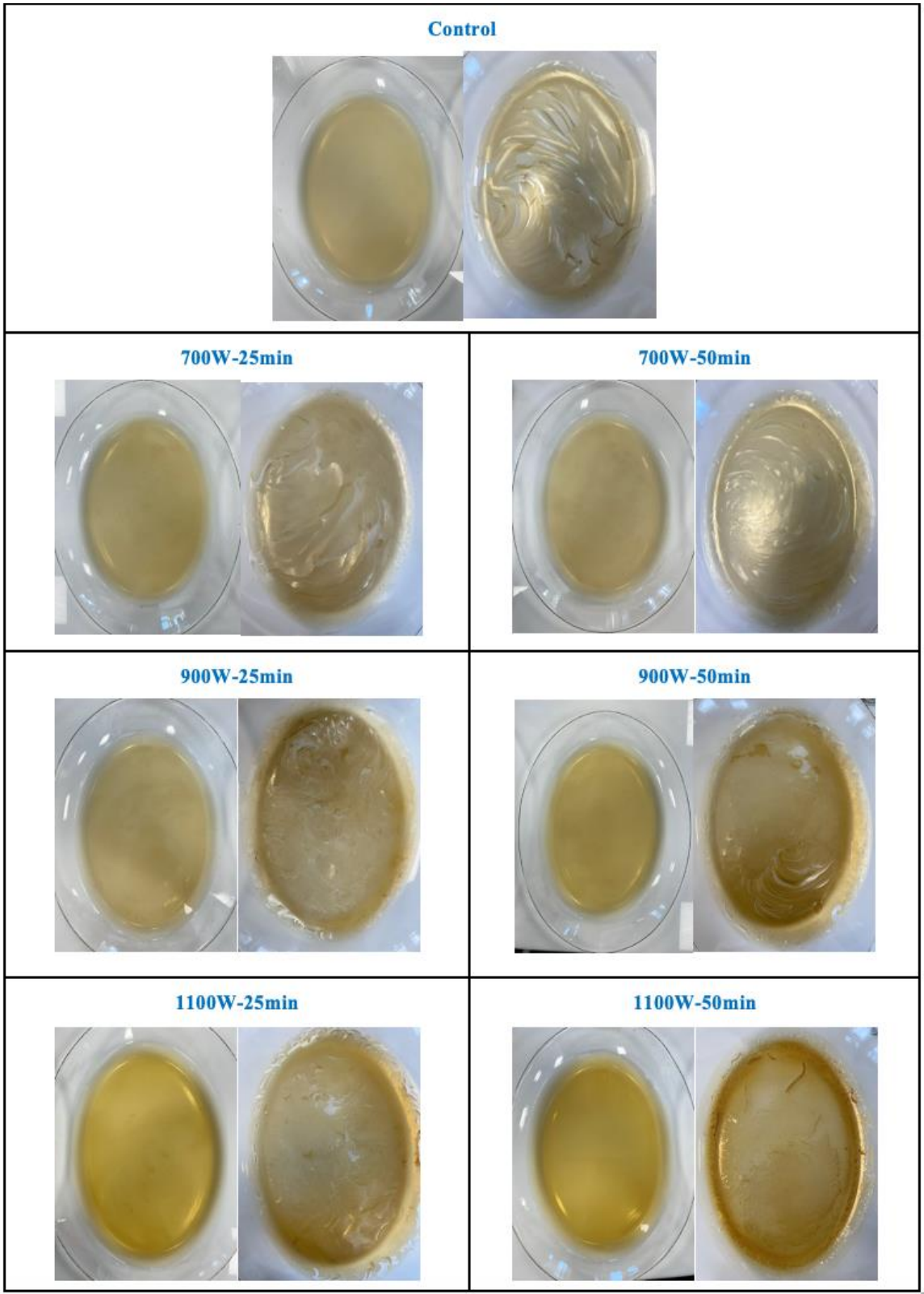
For extraction of mucilage from chia seeds, the method described by Hernández (2012) was modified. Seed:water ratio of 1:20, extraction temperature of 80°C, and extraction time of 2 hours was used. Distilled water heated to 80°C was added to chia seed (50g, dw) in glass bottle (1L) and extracted in a shaking water bath at 80°C for 2 hours. Extraction was finalized by cooling the bottles under cold water at the end of the extraction time (2h). The aqueous suspension was divided into two equal portions and



transferred into two different beakers. As mucilage forms a gel-like structure, an ultrasonication probe (Microson Ultrasonic Cell Disruptor, USA) operating at 14 watts was used to remove the seed from the mucilage gently. Ultrasonication probe was immersed into the suspension to a distance of 6.5cm from the upper level. Ultrasonication was carried out for 3 minutes. Total ultrasonication time of 9 minutes (3 minutes x 3 times) was applied to each sample. Some of the mucilage adhered to the seed was removed by ultrasonication. The liquid portion representing the mucilage was filtered to a glass container. Since the liquid level decreased after removing mucilage, the remaining suspension was transferred to a small beaker in order to adjust the immersing distance of probe to 6.5cm. In preliminary studies, mucilage yield was approximately 6% (dw). In order to obtain approximately 30g of mucilage (amount necessary for all mucilage analysis) for each sample, the extraction process was carried out as 11 replicates by using 50g (dw) of chia seeds at each time for each sample. Mucilage was then dried in an oven at 50°C for 22 hours. Chia mucilages obtained for each infrared power and time treatment before (left) and after (right) drying are given in Figure 12. All 11 replicates of each sample were combined in a large batch and ground by using a mortar and sieved through a 425µm sieve. Finally, the ground samples were stored at +4°C until analysis.

The mucilage yield was calculated by using the formula given below.

$$\text{Mucilage yield (\%, dw)} = \frac{\text{Weight of mucilage (g, dw)}}{\text{Weight of chia seed (g, dw)}} \times 100$$



**Figure 12.** Mucilage samples before and after drying are given on the left and right side of the figure, respectively

### **3.4.1. Analyses of Mucilage Samples**

#### **3.4.1.1. Mucilage Composition**

Moisture contents of mucilage samples (<425 $\mu$ ) were determined at 105°C by using AACC 44-15A (Air oven methods) (AACC, 1983). Ash content of mucilage samples (<425 $\mu$ ) were determined by using AACC Method No 08-01 (AACC, 1990). Protein content (Nx6.25) of mucilage samples (<425 $\mu$ ) were determined by using Kjeldahl method (AACC Method 46-12) (AACC, 1990). The results were given as the average of two values.

The total dietary fiber content of the mucilage samples was determined by using Megazyme Total Dietary Fiber Kit (Megazyme International Ireland Ltd., Ireland). Total dietary fiber method (Section 3.3.5) used for chia samples was also applied for mucilage samples. The results were given as the average of two values.

#### **3.4.1.2. FTIR Spectroscopy**

FTIR spectrums of mucilage samples extracted from the control and infrared treated chia samples were obtained by using Attenuated Total Reflectance-Fourier Transform Infrared (ATR-FTIR) (Thermo Scientific, Nicolet™ iS50, United States) at HUNITEK (Hacettepe Üniversitesi İleri Teknolojiler Uygulama ve Araştırma Merkezi, Ankara, Turkey). Analysis was carried out as 16 scanning frequencies over a wavenumber range of 4000-400  $\text{cm}^{-1}$  with 4  $\text{cm}^{-1}$  resolution (Punia and Dhull, 2019).

#### **3.4.1.3. Functional Properties of Mucilage Samples**

Water holding capacity (Section 3.3.10.1), oil holding capacity (Section 3.3.10.2), emulsion activity and emulsion stability (Section 3.3.10.3) methods used for chia samples were also used for mucilage samples. The results were given as the average of three values. Viscosity of the mucilage samples in 1% and 2% (w/v, dw) solutions was determined by using Rheometer (Brookfield Rheometer, RST CC, USA) with shear rate 400-1400 $\text{s}^{-1}$ . The results were given as the average of two values.

### **3.5. Statistical Analysis**

The results obtained in the present study were evaluated by using one-way analysis of variance (ANOVA) (SPSS 23.0, ABD). When significant differences were found, Duncan comparison test was applied to determine the differences between means. The standard deviation values for some results (protein, ash, color, total dietary fiber, total

phenolic content, total flavonoid content, antioxidant activity, phenolic profiles, functional properties) were calculated by using MS Excel (MS Excel, Microsoft Corp., USA).

## 4. RESULTS AND DISCUSSION

### 4.1. Properties of Infrared Treated Chia Samples

#### 4.1.1. Chemical Composition of Infrared Treated Chia Samples

Moisture, protein, and ash contents of control and infrared treated chia samples are given in Table 2. The moisture contents of the chia samples ranged between 0.49-6.89%. Control samples had the highest moisture content (6.89%). Increase in infrared power and time caused a decrease in the moisture content of the samples. In a study by Hatamian et al. (2020), chia seeds were roasted at different temperatures (160°C-180°C) and times (15-35 minutes). The moisture content of the chia sample decreased from 6.6% to 1%.

**Table 2.** Chemical composition of infrared treated chia samples\*

Sample	Moisture (%)	Protein (% <i>,dw</i> )	Ash (% <i>,dw</i> )
Control	6.89 ± 0.063	22.42 ± 0.097 <sup>a</sup>	4.76 ± 0.006 <sup>c</sup>
700W-25 min	3.23 ± 0.009	21.77 ± 0.558 <sup>a</sup>	4.77 ± 0.008 <sup>b</sup>
700W-50 min	1.53 ± 0.028	21.76 ± 0.145 <sup>a</sup>	4.73 ± 0.000 <sup>d</sup>
900W-25 min	1.17 ± 0.005	21.62 ± 0.245 <sup>a</sup>	4.72 ± 0.001 <sup>d</sup>
900W-50 min	1.05 ± 0.038	21.33 ± 0.008 <sup>a</sup>	4.80 ± 0.009 <sup>a</sup>
1100W-25 min	0.51 ± 0.019	21.73 ± 0.852 <sup>a</sup>	4.75 ± 0.006 <sup>c</sup>
1100W-50 min	0.49 ± 0.010	21.36 ± 0.638 <sup>a</sup>	4.76 ± 0.005 <sup>bc</sup>

\*Values followed by the same letter in the same column are not significantly different ( $\alpha=0.05$ )  
Values are the means of two replicates.

The protein content of the control chia sample used in this study was found to be 22.42% (*dw*). The protein content of the infrared treated chia samples were between 21.33-21.77% (*dw*). The protein content decreased slightly as compared to control. But the decrease was insignificant. In literature, the protein content of chia seeds was reported as 18.3% (Coelho and Salas-Mellado, 2014), 17-20% (Ayerza and Coates, 2005), and 24% (Segura-Campos et al. 2014). In a study by Ghafoor et al. (2018), chia seeds were roasted at 120°C for 20 minutes and it was reported that the protein content of the roasted seed (20.38%) was lower than that of the unroasted seeds (21.81%).

A slight significant difference was observed in ash contents of chia samples. In literature, ash content of chia seeds was reported as 3.63-4.05% (Sargi et al., 2013; Segura-Campos et al., 2014).

#### 4.1.2. Color Values of Infrared Treated Chia Samples

L\*, a\*, b\* color values of the control and infrared treated chia samples are given in Table 3. L\*, a\*, and b\* color values of chia samples were between 42.65-44.50, 1.26-1.75, and 2.15-2.98, respectively. Infrared treatment caused a slight decrease in L\* color values of the chia samples, indicating a slightly dark color. Infrared treatment did not cause a substantial difference in the a\* and b\* color values of the chia samples.

**Table 3.** Color values of infrared treated chia samples

Sample	L*	a*	b*
Control	44.50 ± 0.632	1.52 ± 0.025	2.84 ± 0.042
700W- 25 min	44.30 ± 0.285	1.30 ± 0.061	2.67 ± 0.025
700W-50 min	43.46 ± 0.051	1.26 ± 0.036	2.50 ± 0.021
900W-25 min	43.47 ± 0.284	1.47 ± 0.020	2.25 ± 0.052
900W-50 min	43.33 ± 0.497	1.75 ± 0.047	2.24 ± 0.059
1100W-25 min	43.04 ± 0.616	1.27 ± 0.057	2.15 ± 0.061
1100W-50 min	42.65 ± 0.272	1.60 ± 0.010	2.98 ± 0.021

Values are the means of three replicates.

#### 4.1.3. Total Dietary Fiber Content of Infrared Treated Chia Samples

Total dietary fiber contents (% dw) of the defatted control and infrared treated chia samples are given in Table 4. The total dietary fiber content of control chia sample was found as 36.61 % (dw). In literature, the total dietary fiber content of chia was reported as 39.94-41.41 g/100g seed (dw) for cv. Jalisco and 36.97-38.79 g/100g seed (dw) for cv. Sinaloa (Reyes-Caudillo et al., 2008), and 374.4 g/kg seed (Scapin et al., 2016). The total dietary fiber content of chia sample was higher than that of wheat (11.6-17%, dw), oat (13.7-30.1%, dw), barley (16.8-27.9%, dw), rye (15.2-20.9%, dw), quinoa (16.2-21.6%, dw), and corn (13.1-19.6%, dw) (Vithana Pathirannehelage and Joye, 2020).

In the present study, the total dietary fiber contents of the infrared treated chia samples were between 32.39-40.83% (dw). The total dietary fiber contents of the infrared treated chia samples were significantly higher than that of the control sample, except chia samples infrared treated at 1100W. Among all samples, the highest total dietary fiber content (40.83%, dw) was obtained for 900W-25 min chia sample. Similar increase in total dietary fiber content was also reported for infrared treated soybean samples (Yalcin, 2011).

**Table 4.** Total dietary fiber contents of infrared treated chia samples\*

Sample	Total Dietary Fiber (% dw)
Control	36.61 ± 0.121 <sup>e</sup>
700W- 25 min	37.07 ± 0.118 <sup>d</sup>
700W-50 min	38.94 ± 0.177 <sup>c</sup>
900W-25 min	40.83 ± 0.293 <sup>a</sup>
900W-50 min	39.95 ± 0.059 <sup>b</sup>
1100W-25 min	32.39 ± 0.030 <sup>f</sup>
1100W-50 min	32.68 ± 0.059 <sup>f</sup>

\*Values followed by the same letter are not significantly different ( $\alpha=0.05$ )  
Values are the means of two replicates.

#### 4.1.4. Total Phenolic Content of Infrared Treated Chia Samples

Total phenolic content of the control and infrared treated chia samples were between 2.059-2.376 mg GAE/g dry sample, as shown in Table 5. The total phenolic contents of the infrared treated chia samples were significantly higher than that of the control sample, except 700W-25 min chia sample. When the total phenolic content of the chia samples were evaluated for each infrared treatment time (25 min or 50 min), it was observed that total phenolic content increased significantly as the infrared power increased. For each infrared power, total phenolic content increased significantly as infrared treatment time increased from 25 min to 50 min. Among all samples, the highest total phenolic content (2.376 mg GAE/g dry sample) was obtained for 1100W-50 min chia sample. Similar increase by using infrared treatment was also observed for total phenolic content of soybean samples (Yalcin, 2011).

**Table 5.** Total phenolic content of infrared treated chia samples\*

Sample	Total Phenolic Content (mg gallic acid (GAE)/g dry sample)
Control	2.059 ± 0.0091 <sup>e</sup>
700W- 25 min	2.065 ± 0.0127 <sup>e</sup>
700W-50 min	2.097 ± 0.0134 <sup>d</sup>
900W-25 min	2.205 ± 0.0207 <sup>c</sup>
900W-50 min	2.261 ± 0.0142 <sup>b</sup>
1100W-25 min	2.262 ± 0.0069 <sup>b</sup>
1100W-50 min	2.376 ± 0.0231 <sup>a</sup>

\*Values followed by the same letter are not significantly different ( $\alpha=0.05$ )  
Values are the means of four replicates.

In literature, the total phenolic contents of chia seeds were reported as 0.92 mg GAE/g (cv. Jalisco) and 0.88 mg GAE/g (cv. Sinaloa) (Reyes-Caudillo et al., 2008), 0.94 mg GAE/g (Marineli et al., 2014), 97.7 mg GAE/100 g (dw) (Beltrán-Orozco et al., 2020),

1.6398 mg GAE/g (Martínez-Cruz and Paredes-López, 2014), 1.65 mg GAE/g dry sample (Khursheed et al., 2023), 2.14-2.21 mg GAE/g (white chia seeds), and 2.18-2.19 mg GAE/g (dark chia seeds) (Alvites-Misajela et al., 2019).

In a study by Haripriya (2020), the total phenolic content of chia seed increased from 43.19 mg/100g TAE to 45.79 mg/100g TAE by roasting in a griddle for 5-7 min. In a study by Hatamian et al. (2020), roasting at 160°C caused an increase (for 15 min 1.9 mg GAE/g; for 25 min 2.7 mg GAE/g and for 35 min 2.1 mg GAE/g) in total phenolic content as compared to control (1.6 mg GAE/g). Although a decrease was reported for roasting at 180°C for 15 min (1.5 mg GAE/g) and 25 min (1.1 mg GAE/g), the highest total phenolic content was stated for the sample roasted for 35 min (3.2 mg GAE/g). Ghafoor et al. (2018) reported that the total phenolic contents of chia seeds increased from 3.07 mg GAE/g to 3.43 mg GAE/g by roasting 120°C for 20 minutes. Ghafoor et al. (2022) also reported an increase (from 349.44 mg GAE/100g to 535 mg GAE/100g) in total phenolic contents of chia seeds by roasting at 120°C for 6 minutes in a teflon pan. Similar to the results obtained in the present study, heat treatment caused an increase in the total phenolic content of the chia samples. The increase in total phenolic content of heat treated chia seeds could be due to the improved extractability of bound phenolics through the thermal breakdown of cellular components (Haripriya, 2020). However, a decrease (from 0.98 mg GAE/100g to 0.51mg GAE/100g) in total phenolic content was reported for chia seeds roasted at different temperatures (90, 120, 150, and 180 °C) (Ghafoor et al., 2020).

#### **4.1.5. Total Flavonoid Content of Infrared Treated Chia Samples**

Total flavonoid contents of the control and infrared treated chia samples are given in Table 6. The total flavonoid content of the control sample was 1.157 mg quercetin (QE)/g dry sample. Infrared treatment at 700W or 900W for 25 min or 50 min resulted an increase in total flavonoid content of the chia samples. The increase in total flavonoid content was only significant for 700W-50 min chia sample. Among the samples, the highest flavonoid content (1.285 mg quercetin (QE)/g dry sample) was obtained for 700W-50 min chia sample.

Improved extractability of flavonoids through the thermal breakdown of cellular components may lead to an increase in total flavonoid content of chia seeds. However, infrared treatment at higher power (1100W for 25 min or 50 min) caused a decrease in total flavonoid content.



**Table 6.** Total flavonoid content of infrared treated chia samples\*

Sample	Total Flavonoid Content (mg quercetin (QE) / g dry sample)
Control	1.157± 0.0014 <sup>bc</sup>
700W- 25 min	1.174 ± 0.0482 <sup>bc</sup>
700W-50 min	1.285 ± 0.0549 <sup>a</sup>
900W-25 min	1.212 ± 0.0331 <sup>b</sup>
900W-50 min	1.190 ± 0.0073 <sup>b</sup>
1100W-25 min	1.147 ± 0.0159 <sup>bc</sup>
1100W-50 min	1.121 ± 0.0342 <sup>c</sup>

\*Values followed by the same letter are not significantly different ( $\alpha=0.05$ )

Values are the means of three replicates.

In literature, the total flavonoid contents of chia seeds were reported as 1.50-1.56 mg CE/g (white chia seeds), 1.54-1.57 mg CE/ g (dark chia seeds) (Alvites-Misajela et al., 2019), 0.35 mg QE/100 g dry sample (Khursheed et al., 2023), and 35.8 mg QE/100 g (dw) (Beltrán-Orozco et al., 2020). Scapin et al. (2016) extracted flavonoids of chia seeds at different temperatures (40, 50, 60°C). The total flavonoid content was reported as 0.110-0.162 g/kg QE.

In a study by Ghafoor et al. (2018), as compared to total flavonoid content of control (2.21 mg CE/g), higher total flavonoid content for chia seeds roasted in oven at 120°C-20 minutes (2.51 mg CE/g) was reported. A gradual decrease (from 17.62 mg CE/100 g to 9.86 mg CE/100 g) in total flavonoid content was reported by Ghafoor et al. (2020) for roasting chia seeds at 90, 120, 150, and 180 °C. Similarly, a decrease (from 1217.62 mg QE /100g to 1020.48 mg QE /100g) in total flavonoid content was reported by Ghafoor et al. (2022) chia seeds roasted at 120°C for 6 minutes in a teflon pan.

#### 4.1.6. Antioxidant Activity of Infrared Treated Chia Samples

Antioxidant activities of control and infrared treated chia samples were determined by DPPH (% radical scavenging activity) and QUENCHER-CUPRAC methods.

##### 4.1.6.1. Radical scavenging activity (DPPH)

The radical scavenging activities (% , DPPH) of the chia samples are presented in Table 7. The radical scavenging activities of control and infrared treated chia samples were between 75.11-80.71%. Infrared treatment caused a significant increase in radical scavenging activities of chia samples. Significantly highest (80.71%) radical scavenging activity was obtained for 1100W-50 min chia sample.

When the radical scavenging activities of the chia samples were evaluated for each infrared treatment time (25 min or 50 min), it was observed that radical scavenging activities increased significantly as the infrared power increased. For each infrared power, radical scavenging activities significantly increased as infrared treatment time increased from 25 min to 50 min.

**Table 7.** Radical scavenging activity (%) of infrared treated chia samples\*

Sample	Radical Scavenging Activity (%)
Control	75.11 ± 0.153 <sup>f</sup>
700W-25 min	76.79 ± 0.305 <sup>e</sup>
700W-50 min	77.66 ± 0.176 <sup>d</sup>
900W-25 min	78.63 ± 0.153 <sup>c</sup>
900W-50 min	80.00 ± 0.153 <sup>b</sup>
1100W-25 min	80.15 ± 0.153 <sup>b</sup>
1100W-50 min	80.71 ± 0.176 <sup>a</sup>

\*Values followed by the same letter are not significantly different ( $\alpha=0.05$ )

Values are the means of three replicates.

In literature, the radical scavenging activities (% , DPPH) of chia seeds were reported as 30% (Hatamian et al., 2020), 68.83% (Martínez-Cruz and Paredes-López, 2014) and 88.27% (Ghafoor et al., 2020). It was reported that high antioxidant activity of chia was due to the presence of phenolic substances such as phenolic acids, isoflavones, and anthocyanins (Martínez-Cruz and Paredes-López, 2014).

Ghafoor et al. (2020) investigated the effect of roasting (90, 120, 150, and 180°C) on the antioxidant activity (% , DPPH) of chia seeds, roasted in oven at different temperatures. Antioxidant activity of control chia seed was reported as 88.27%. Antioxidant activity decreased to 85.76% at 90°C, 79.57% at 120°C, 48.44% at 150°C, and 31.23% at 180°C. In contrast to the decrease reported by Ghafoor et al. (2020), an increase from 75.11% to 80.71% was observed by infrared treatment of chia seeds in the present study.

In a study by Hatamian et al. (2020), an increase in antioxidant activity (% , DPPH) was observed by roasting the chia seeds at 160°C and 180°C for 15 min, 25 min, and 35 min. Antioxidant activity of control chia was reported as 30%. The highest antioxidant activity was reported as 61%, for the chia samples roasted at 180°C for 25 min.

#### 4.1.6.2. QUENCHER-CUPRAC

Total antioxidant capacity (TAC) values of the chia samples are given in Table 8. The trolox calibration curve, which was used to calculate the TAC values of the samples, is

shown in Figure 5. The TAC values of the infrared treated chia seeds were significantly higher than that of the control. Among all chia samples, significantly highest (211.63 mmol TE/kg sample) TAC value was obtained for 1100W-50 min chia sample.

When the total antioxidant capacity (TAC) of the chia samples were evaluated for each infrared treatment time (25 min or 50 min), it was observed that total antioxidant capacity significantly increased as the infrared power increased. For each infrared power, total antioxidant capacity significantly increased as infrared treatment time increased from 25 min to 50 min.

**Table 8.** Total antioxidant capacity (TAC (mmol Trolox/kg sample) of the infrared treated chia samples\*

Sample	Total Antioxidant Capacity (TAC, mmol TE/kg sample)
Control	83.89 ± 0.448 <sup>g</sup>
700W-25 min	118.24 ± 0.224 <sup>f</sup>
700W-50 min	124.57 ± 0.224 <sup>e</sup>
900W-25 min	137.39 ± 0.448 <sup>d</sup>
900W-50 min	178.39 ± 0.224 <sup>c</sup>
1100W-25 min	193.27 ± 0.224 <sup>b</sup>
1100W-50 min	211.63 ± 0.672 <sup>a</sup>

\*Values followed by the same letter are not significantly different ( $\alpha=0.05$ )  
Values are the means of two replicates.

Tufan et al. (2012) determined the total antioxidant capacity of some cereals by using the same method. TAC values (mmol TE kg<sup>-1</sup>) of the cereal samples were reported as follows; wheat germ (40.22±1.86) > barley (26.35±0.61) > rye (16.21±0.25) > sorghum (13.55±0.55) > wheat (13.44±0.46) > oat (10.46±0.23). In the present study, higher values for total antioxidant capacity by using QUENCHER-CUPRAC method were obtained for chia samples as compared to those of different cereals, reported by Tufan et al. (2012).

#### 4.1.7. Phenolic Profile of Infrared Treated Chia Samples

Some phenolic and flavonoid compounds determined by HPLC analysis of the methanolic extracts of chia seeds are given in Table 9. Standard curves of caffeic acid, chlorogenic acid, p-Coumaric acid, ferulic acid, rutin, and quercetin are given in Figures 6-11. Infrared treatment caused a significant decrease in caffeic acid and p-Coumaric acid and caused a significant increase in chlorogenic acid, ferulic acid (except 1100W-50 min), and rutin (not detected for 700W-25 min chia). Infrared treatment caused an increase only in quercetin content of 700W-50 min and 900W-25 min chia samples. Improved extractability of phenolics through the thermal breakdown of cellular components by the aid of infrared may lead to an increase in phenolic compounds (chlorogenic acid, ferulic acid, rutin, quercetin) in chia seeds.

Caffeic acid content of control chia sample (0.010 mg/g (dw)) was significantly higher as compared to infrared treated chia samples. When the caffeic acid contents of the chia samples were evaluated for each infrared treatment time (25 min or 50 min), it was observed that the amount of caffeic acid decreased significantly as the infrared power increased, except for 1100W-50 min chia sample. The caffeic acid was not detected in the 1100W-50 min chia sample. The amount of caffeic acid did not change significantly as the infrared treatment time increased from 25 min to 50 min, for infrared treatment at 700W and 900W. In literature, caffeic acid content of chia seeds were reported as 0.45 mg/100 g (dw) (Pająk et al., 2019), 0.0030 mg/g seed (cv. Jalisco) and 0.00680 mg/g seed (cv. Sinaloa) (Reyes-Caudillo et al., 2008), 30.89 µg/g (Coelho and Salas-Mellado, 2014), 31.14 mg/kg (Ghafoor et al., 2018), 1.75 mg/100g (Ghafoor et al., 2022), and 0.280 mg/g (dw) (Abdel-Aty et al., 2021). When the caffeic acid content of the chia sample used in the present study was compared with the ones reported above, higher caffeic acid content was only reported by Abdel-Aty et al. (2021).

Among all chia samples, highest (0.053 mg/g (dw)) p-Coumaric acid content was observed in the control chia sample. For each infrared treatment time (25 min or 50 min), as the infrared power increased, the amount of p-Coumaric acid decreased significantly, except for 1100W-50 min chia sample. p-Coumaric acid was not detected in the 1100W-50min chia sample. In literature, p-Coumaric acid content of chia seeds were reported as 2.93 mg/100g (Ghafoor et al., 2022) and 4.17 mg /kg (Ghafoor et al., 2018). The results of our study for p-Coumaric acid were higher than that reported by Ghafoor et al. (2018) and Ghafoor et al. (2022).

Chlorogenic acid content of control chia sample was found to be 0.024 mg/g (dw). Chlorogenic acid significantly increased as the infrared power and time increased. The highest (0.118 mg/g (dw)) chlorogenic acid content was observed in the 1100W-50 min chia sample. In literature, chlorogenic acid contents of the chia seeds were reported as 4.68 µg/g (Coelho and Salas-Mellado, 2014), 0.028 mg/g (dw) (Abdel-Aty et al., 2021), 0.102 mg/g seed (cv. Jalisco) and 0.0459 mg/g seed (cv. Sinaloa) (Reyes-Caudillo et al., 2008). Chlorogenic acid content found in the present study was higher as compared to that reported by Coelho and Salas-Mellado (2014).

Ferulic acid was not detected in the control chia sample. Ferulic acid of chia sample generally increased by infrared treatment, except for 1100W-50 min treatment. The highest (0.021 mg/g (dw)) ferulic acid content was observed in the 1100W-25 min chia sample. When the ferulic acid contents of the chia samples were evaluated for each infrared treatment time (25 minutes or 50 minutes), it was observed that the ferulic acid content increased significantly as the infrared power increased, except for 1100W for 50 min treatment. In literature, ferulic acid contents of chia seeds were reported as 2.14 mg/kg (Ghafoor et al., 2018), 0.16 mg/100 g (dw) (Pająk et al., 2019), 0.54 mg/100g (Ghafoor et al., 2022), and 0.112 mg/g (dw) (Abdel-Aty et al., 2021).

Rutin was not detected in the control and 700W-25 min chia sample. Extractability of rutin was initially observed at 700W-50 min sample by the aid of infrared treatment. The highest (0.265 mg/g (dw)) rutin content was observed in the 1100W for 50 min chia sample. The amount of rutin increased significantly as the infrared treatment time increased from 25 min to 50 min, for infrared treatment at 900W and 1100W. Ghafoor et al. (2022) reported rutin content of chia seeds as 3.98 mg/100g.

Quercetin is the mostly known flavonoid in chia samples. Quercetin content of control chia sample was found to be 0.206 mg/g (dw). The highest (0.211 mg /g (dw)) quercetin content was found in the 700W-50 min chia sample. Quercetin content (0.208 mg/g (dw)) of 900W-25 min sample was not significantly different than that of 700W-50 min sample. As compared to control, 900W-50 min, 1100W-25 min, and 1100W-50 min samples had significantly lower quercetin contents. In literature, quercetin contents of chia seeds were reported as 0.17 µg/g (Coelho and Salas-Mellado, 2014), 0.080 mg/g (dw) (Abdel-Aty et al., 2021), and 5.01 mg/100g (Ghafoor et al., 2022). Higher quercetin content was found for chia in the present study.

**Table 9.** Phenolic profile of infrared treated chia samples (mg/g dw)\*

Sample	Caffeic acid	p-Coumaric acid	Chlorogenic acid	Ferulic acid	Rutin	Quercetin
Control	0.010 ± 0.0001 <sup>a</sup>	0.053 ± 0.0024 <sup>a</sup>	0.024 ± 0.0002 <sup>g</sup>	ND	ND	0.206 ± 0.0029 <sup>bc</sup>
700W-25 min	0.008 ± 0.0002 <sup>b</sup>	0.046 ± 0.0007 <sup>b</sup>	0.038 ± 0.0007 <sup>f</sup>	0.008 ± 0.0002 <sup>e</sup>	ND	0.204 ± 0.0041 <sup>c</sup>
700W-50 min	0.009 ± 0.0001 <sup>b</sup>	0.045 ± 0.0002 <sup>b</sup>	0.047 ± 0.0006 <sup>e</sup>	0.010 ± 0.0008 <sup>d</sup>	0.039 ± 0.0009 <sup>e</sup>	0.211 ± 0.0035 <sup>a</sup>
900W-25 min	0.007 ± 0.0004 <sup>c</sup>	0.027 ± 0.0002 <sup>c</sup>	0.094 ± 0.0010 <sup>d</sup>	0.018 ± 0.0012 <sup>b</sup>	0.061 ± 0.0007 <sup>d</sup>	0.208 ± 0.0027 <sup>ab</sup>
900W-50 min	0.007 ± 0.0002 <sup>c</sup>	0.019 ± 0.0002 <sup>d</sup>	0.098 ± 0.0014 <sup>c</sup>	0.019 ± 0.0017 <sup>b</sup>	0.103 ± 0.0036 <sup>c</sup>	0.193 ± 0.0009 <sup>d</sup>
1100W-25 min	0.005 ± 0.0002 <sup>d</sup>	0.006 ± 0.0006 <sup>c</sup>	0.114 ± 0.0010 <sup>b</sup>	0.021 ± 0.0013 <sup>a</sup>	0.153 ± 0.0018 <sup>b</sup>	0.185 ± 0.0016 <sup>e</sup>
1100W-50 min	ND	ND	0.118 ± 0.0003 <sup>a</sup>	0.014 ± 0.0007 <sup>c</sup>	0.265 ± 0.0091 <sup>a</sup>	0.125 ± 0.0003 <sup>f</sup>

ND: Not detected

\*Values followed by the same letter in the same column are not significantly different ( $\alpha=0.05$ )

Values are the means of four replicates.

Ghafoor et al. (2018) reported differences in phenolic components of chia seeds by roasting. Roasting of chia seeds at 120°C for 20 min caused increases in caffeic acid (from 31.14 mg /kg to 35.46 mg/kg), p-Coumaric acid (from 4.17 mg /kg to 5.48 mg/kg), and ferulic acid (from 2.14 mg /kg to 2.86 mg/kg). In contrast to the increase reported by Ghafoor et al. (2018), a decrease in caffeic acid and p-Coumaric acid content was observed for infrared treated samples in the present study. In the present study, higher ferulic acid contents in infrared treated chia seeds were observed as compared to the results reported by Ghafoor et al. (2018) for roasted chia seeds.

In a study by Ghafoor et al. (2022), it was reported that roasting of chia seeds at 120°C for 6 minutes in a teflon pan caused a decrease in caffeic acid (from 1.75 mg/100g to 0.50 mg/100g), p-Coumaric acid (2.93 mg/100g to 1.28 mg/100g), and quercetin (from 5.01 mg/100g to 4.08 mg/100g). The ferulic acid content (0.54 mg/100g) was reported as the same for raw and roasted chia seeds. The amount of rutin in chia seed was reported as 3.98 mg/100g, while it was reported as 24.92 mg/100g in roasted chia seed. The decrease in caffeic and p-Coumaric acid and the increase in rutin content of chia seeds by roasting (Ghafoor et al., 2022) are similar to the ones found in the present study.

#### **4.1.8. Functional Properties of Infrared Treated Chia Seeds**

##### **4.1.8.1. Water and Oil Holding Capacity**

The water holding capacity and oil holding capacity of control and infrared treated chia samples are given in Table 10. The water holding capacity of chia samples were between 1.82-8.44 g water/g dry sample. Among all chia samples, highest (8.44 g water/g dry sample) water holding capacity was obtained for control chia sample. Infrared treatment caused a significant decrease in water holding capacity of chia samples.

When the water holding capacity of the chia samples were evaluated for each infrared treatment time (25 min or 50 min), it was observed that water holding capacity decreased significantly as the infrared power increased. For each infrared power, water holding capacity decreased significantly as infrared treatment time increased from 25 min to 50 min.

Oil holding capacity of chia samples were between 1.72-1.94 g oil/g dry sample. The oil holding capacity of the control sample was significantly higher than that of the infrared treated chia samples. Infrared treatment caused a significant decrease in oil holding capacity of chia samples.

**Table 10.** Water and oil holding capacity of infrared treated chia samples\*

<b>Sample</b>	<b>Water Holding Capacity (g water/g dry sample)</b>	<b>Oil Holding Capacity (g oil/g dry sample)</b>
Control	8.44 ± 0.050 <sup>a</sup>	1.94 ± 0.027 <sup>a</sup>
700W-25 min	7.61 ± 0.093 <sup>b</sup>	1.89 ± 0.028 <sup>b</sup>
700W-50 min	5.37 ± 0.047 <sup>c</sup>	1.85 ± 0.034 <sup>c</sup>
900W-25 min	3.37 ± 0.039 <sup>d</sup>	1.82 ± 0.001 <sup>cd</sup>
900W-50 min	2.30 ± 0.099 <sup>e</sup>	1.80 ± 0.016 <sup>de</sup>
1100W-25 min	2.12 ± 0.014 <sup>f</sup>	1.78 ± 0.022 <sup>e</sup>
1100W-50 min	1.82 ± 0.078 <sup>g</sup>	1.72 ± 0.014 <sup>f</sup>

\*Values followed by the same letter in the same column are not significantly different ( $\alpha=0.05$ )  
Values are the means of three replicates.

In literature, water holding capacity of chia seed was reported as 5 g water/g sample (for whole chia seed), 5.5 g water/g sample (for chia seed powder) (Darwish et al., 2018), 6g/100g (Hatamian et al., 2020), 10.64 g/g (solvent meal) and 10.58 g/g (pressed meal) (Capitani et al., 2012).

In literature, oil holding capacity of chia seed was reported as 2.03 g/g (solvent meal) and 1.26 g/g (pressed meal) (Capitani et al., 2012), 3.9 g/100g (Hatamian et al., 2020), 2.23 ml/g (García-Salcedo et al., 2018), 2.7 ml/g (Haripriya, 2020), 3.25 g oil/g sample (for whole chia seed), 2.5 g oil/g sample (for chia seed powder) (Darwish et al., 2018).

Similar to our results, a decrease in water absorption and oil absorption capacity was also reported for roasting of chia seeds in a griddle for 5-7 min (Haripriya, 2020). The water and oil absorption of control chia was reported as 5.2 ml/g and 1.8 ml/g, respectively. By roasting of chia seeds, the water and oil absorption decreased to 2.7 ml/g and 1.2 ml/g, respectively (Haripriya, 2020). In contrast to the decrease for water absorption capacity and oil absorption capacity by infrared treatment in the present study, increases by roasting were reported by Hatamian et al. (2020). Water absorption capacities of chia samples roasted at 185°C for 25 min (6.3 g/100g) and for 35 min (6.4 g/100g) were higher as compared to that of control (6 g/100g). Oil absorption capacity (5.3 g/100g) of roasted (185°C for 35 min) chia sample was higher as compared to that of control (3.9 g/100g) (Hatamian et al., 2020).

The ability of food protein to bind water and oil is influenced by internal factors such as the composition of amino acids, the shape of the protein and the degree of polarity or hydrophobicity on the surface. The water absorption capacity is strongly associated with the quantity of amino acids found in various types of chia and the presence of available



protein functional groups within the chia seed (Haripriya, 2020). Alfredo et al. (2009) reported that the fiber structure and high proportions of hemicellulose and lignin may contribute to the high water holding capacity of chia seeds. The FTIR spectrums of the chia samples are presented in detail in Section 4.1.9. According to the FTIR spectrums intensity of the peak at  $1636\text{ cm}^{-1}$  increased in the samples infrared treated at 700W and 900W for 25 and 50 min as compared to that of control. However, the intensity of this peak decreased in the chia samples infrared treated at 1100W for 25 and 50 min. Hatamian et al. (2020) reported that the change in the peak at  $1600\text{ cm}^{-1}$  corresponding to Amide I group, might indicate the impact of roasting of chia on amides, amino acids, aldehydes, and esters (Hatamian et al., 2020). As discussed above, the changes in FTIR spectrums related to the protein might cause the changes in water holding capacities of infrared treated chia samples.

#### **4.1.8.2. Emulsion Activity and Emulsion Stability**

Emulsion activity and emulsion stability values of control and infrared treated chia samples are given in Table 11. The emulsion activities of the chia samples were between 1.9-22.7%. Among all chia samples, highest emulsion activity (22.7%) was obtained for 700W-50 min chia sample. As compared to control, 700W-25 min, 700W-50 min, and 900W-25 min chia samples gave significantly higher emulsion activity values while 900W-50 min, 1100W-25 min, and 1100W-50 min chia samples gave significantly lower emulsion activity values. Infrared treatment at 700W caused a significant increase in emulsion activity values of chia samples as infrared treatment time increased from 25 min to 50 min. However, for the samples infrared treated at 900W and 1100W, a significant decrease in emulsion activity was observed when the treatment time increased from 25 min to 50 min.

The emulsion stabilities of the chia samples were between 1.3-12%. Among all chia samples, similarly to emulsion activity value, highest emulsion stability (12%) was obtained for 700W-50 min chia sample. As compared to control, 700W-25 min and 700W-50 min chia samples gave significantly higher emulsion stability values while all chia samples infrared treated at 900W or 1100W for 25 min or 50 min gave significantly lower emulsion stability values. As the infrared treatment time increased from 25 min to 50 min, emulsion stability values decreased in chia samples treated at 900W or 1100W but an increase was observed for the samples infrared treated at 700W.

**Table 11.** Emulsion activity and emulsion stability of infrared treated chia samples\*

Sample	Emulsion Activity (%)	Emulsion Stability (%)
Control	20.5 ± 0.22 <sup>d</sup>	9.9 ± 0.18 <sup>c</sup>
700W-25 min	20.7 ± 0.02 <sup>c</sup>	11.3 ± 0.09 <sup>b</sup>
700W-50 min	22.7 ± 0.19 <sup>a</sup>	12.0 ± 0.27 <sup>a</sup>
900W-25 min	22.3 ± 0.09 <sup>b</sup>	8.1 ± 0.18 <sup>d</sup>
900W-50 min	7.0 ± 0.10 <sup>e</sup>	2.9 ± 0.20 <sup>e</sup>
1100W-25 min	2.6 ± 0.10 <sup>f</sup>	1.9 ± 0.10 <sup>f</sup>
1100W-50 min	1.9 ± 0.09 <sup>g</sup>	1.3 ± 0.10 <sup>g</sup>

\*Values followed by the same letter in the same column are not significantly different ( $\alpha=0.05$ )

Values are the means of three replicates.

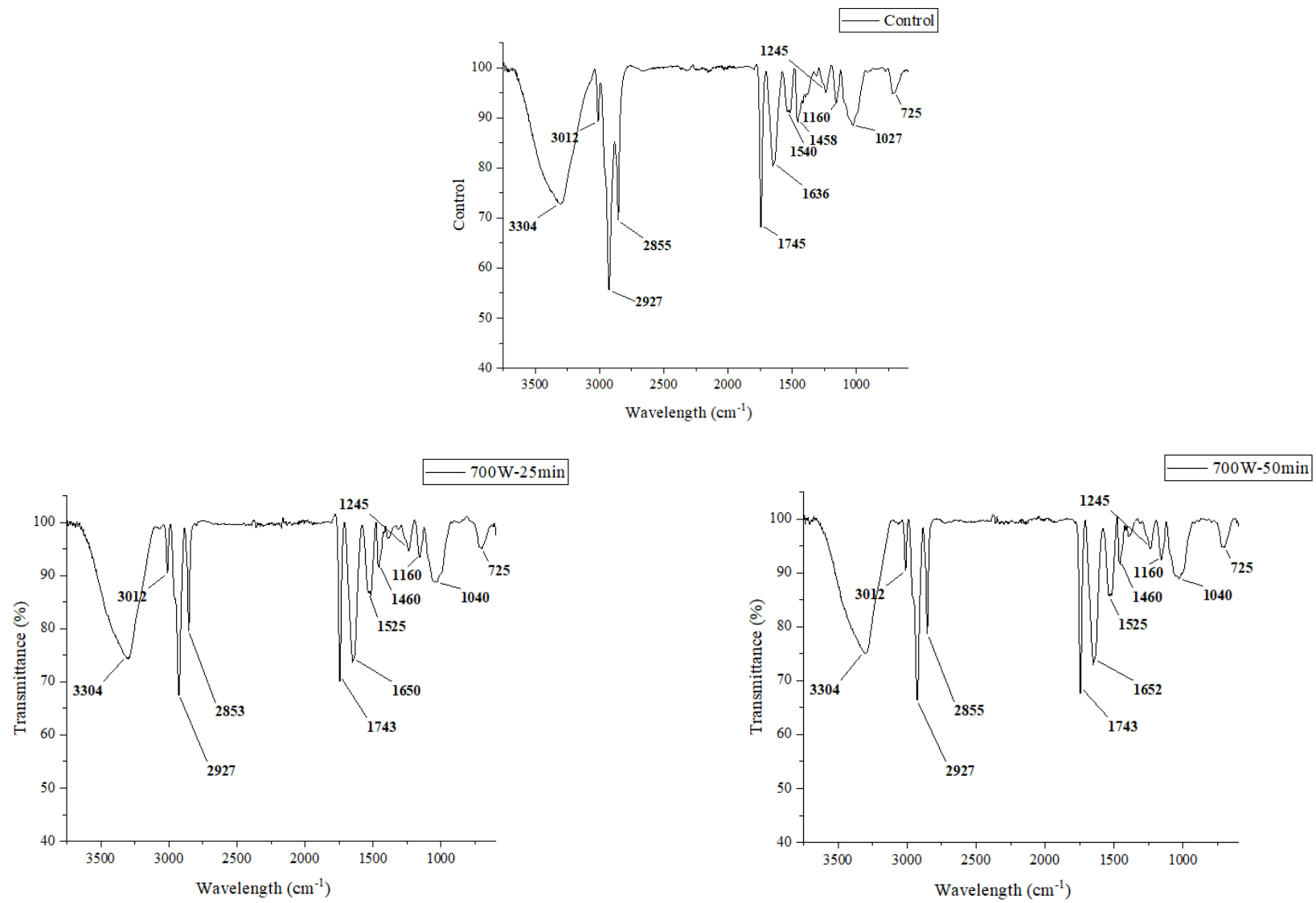
In literature, emulsion activity of chia was reported as 14.69% (García-Salcedo et al., 2018) and 42% (Haripriya, 2020). As compared to the emulsion activity found in the present study, lower emulsion activity value was reported by García-Salcedo et al. (2018) and higher value was reported by Haripriya (2020). Haripriya (2020) reported that the emulsion activity of chia seed decreased from 42% to 38.8% by roasting in a griddle for 5-7 min.

#### 4.1.9. Chemical Structure of Chia Samples (determined by ATR-FTIR)

FTIR spectrums of control and infrared treated chia samples are presented in Figure 13 and Figure 14 (single graph for all samples). The specific peaks given for chia seed in the literature are summarized in Table 12. Figure 13 and Figure 14 showed that all chia samples had similar characteristic absorption peaks given in literature for chia (Table 12). In the present study, specific peaks were observed at wavelengths of 3300-3304  $\text{cm}^{-1}$ , 3012-3015  $\text{cm}^{-1}$ , 2925-2927  $\text{cm}^{-1}$ , 2850-2855  $\text{cm}^{-1}$ , 1740-1745  $\text{cm}^{-1}$ , 1636-1652  $\text{cm}^{-1}$ , 1520-1540  $\text{cm}^{-1}$ , 1458-1460  $\text{cm}^{-1}$ , 1158-1160  $\text{cm}^{-1}$ , 1038-1040  $\text{cm}^{-1}$ , and 725  $\text{cm}^{-1}$ .

In the present study, characteristic absorption peaks of the chia samples observed around 3300  $\text{cm}^{-1}$  could be attributed to OH stretching (Table 12) (Hatamian et al., 2020). The peaks detected around 3012 and 2925  $\text{cm}^{-1}$  could be assigned to C=C-H stretching and C-H stretching (Table 12) (Darwish and El-Sohaimy, 2018; García-Salcedo et al., 2018; Hatamian et al., 2020; Noshad et al., 2020). The peak observed at 2850-2855  $\text{cm}^{-1}$  for chia samples was assigned to C-H stretching (Darwish and El-Sohaimy, 2018; Hatamian et al., 2020; Noshad et al., 2020). Stretching of C-H bonds in methyl groups of chia samples was previously reported at 2872, 2933, 2922  $\text{cm}^{-1}$  by Darwish and El-Sohaimy (2018) and at 2800-3000  $\text{cm}^{-1}$  by Hatamian et al. (2020) and Noshad et al. (2020). The

peak observed around 1740-1745  $\text{cm}^{-1}$  for chia samples could be attributed to C=O stretching (Darwish and El-Sohaimy, 2018; García-Salcedo et al., 2018; Hatamian et al., 2020; Noshad et al., 2020). The peak detected around 1600  $\text{cm}^{-1}$  could be attributed to Amide I groups (Hatamian et al., 2020). The peak around 1500  $\text{cm}^{-1}$  was assigned to -COO- of uronic acids and C=O stretching in uronic acids (Darwish and El-Sohaimy, 2018; Noshad et al., 2020). The peak observed around 1458-1460  $\text{cm}^{-1}$  for chia could be assigned to H-C-H stretching and -COO- of uronic acids (Darwish and El-Sohaimy, 2018; Timilsena et al., 2016). The peak detected at 1245  $\text{cm}^{-1}$  was attributed to the presence of Amide III (Hatamian et al., 2020; Noshad et al., 2020). The peak observed around 1158  $\text{cm}^{-1}$  could be attributed to C-O-C in the pyranose ring (Darwish and El-Sohaimy, 2018). The peak observed at 1038  $\text{cm}^{-1}$  could be attributed to pyranose ring (García-Salcedo et al., 2018). The peak detected at 725  $\text{cm}^{-1}$  was assigned to  $\text{CH}_2$  with more than 7 carbon atoms (Darwish and El-Sohaimy, 2018).



**Figure 13.** FTIR spectrums of control and infrared treated chia samples

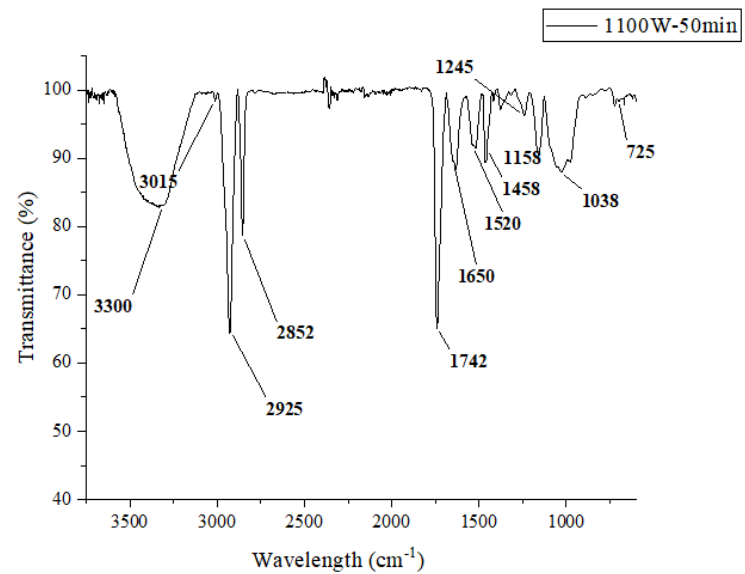
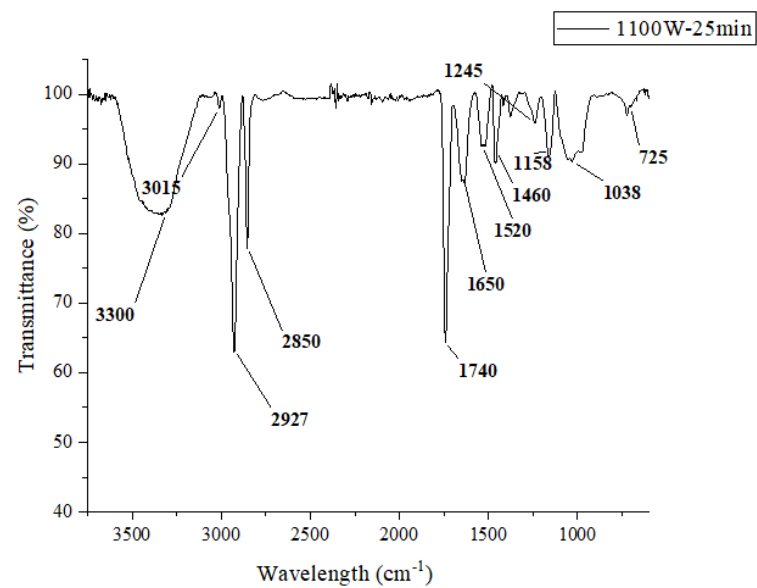
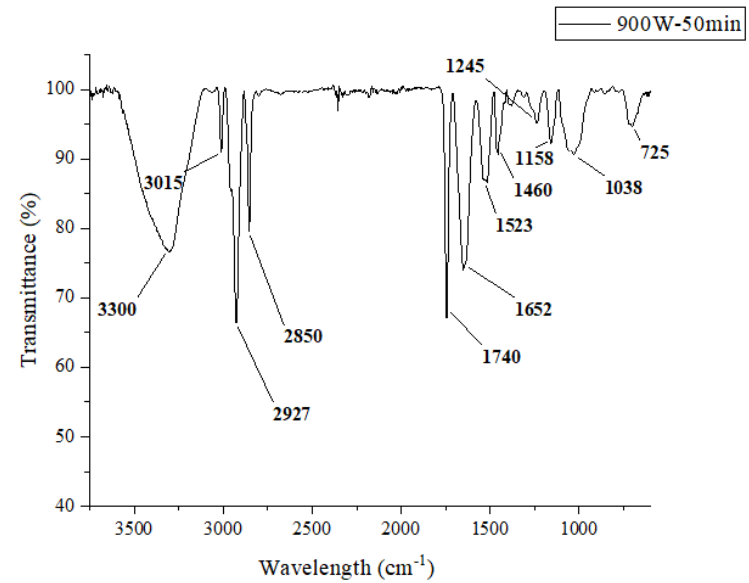
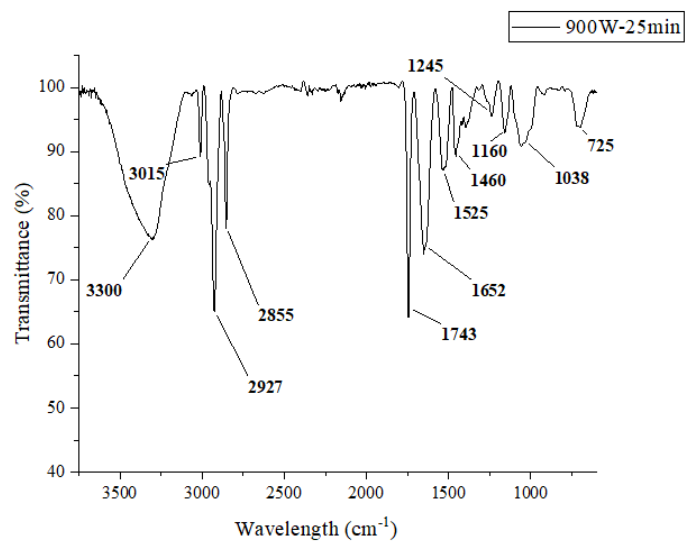
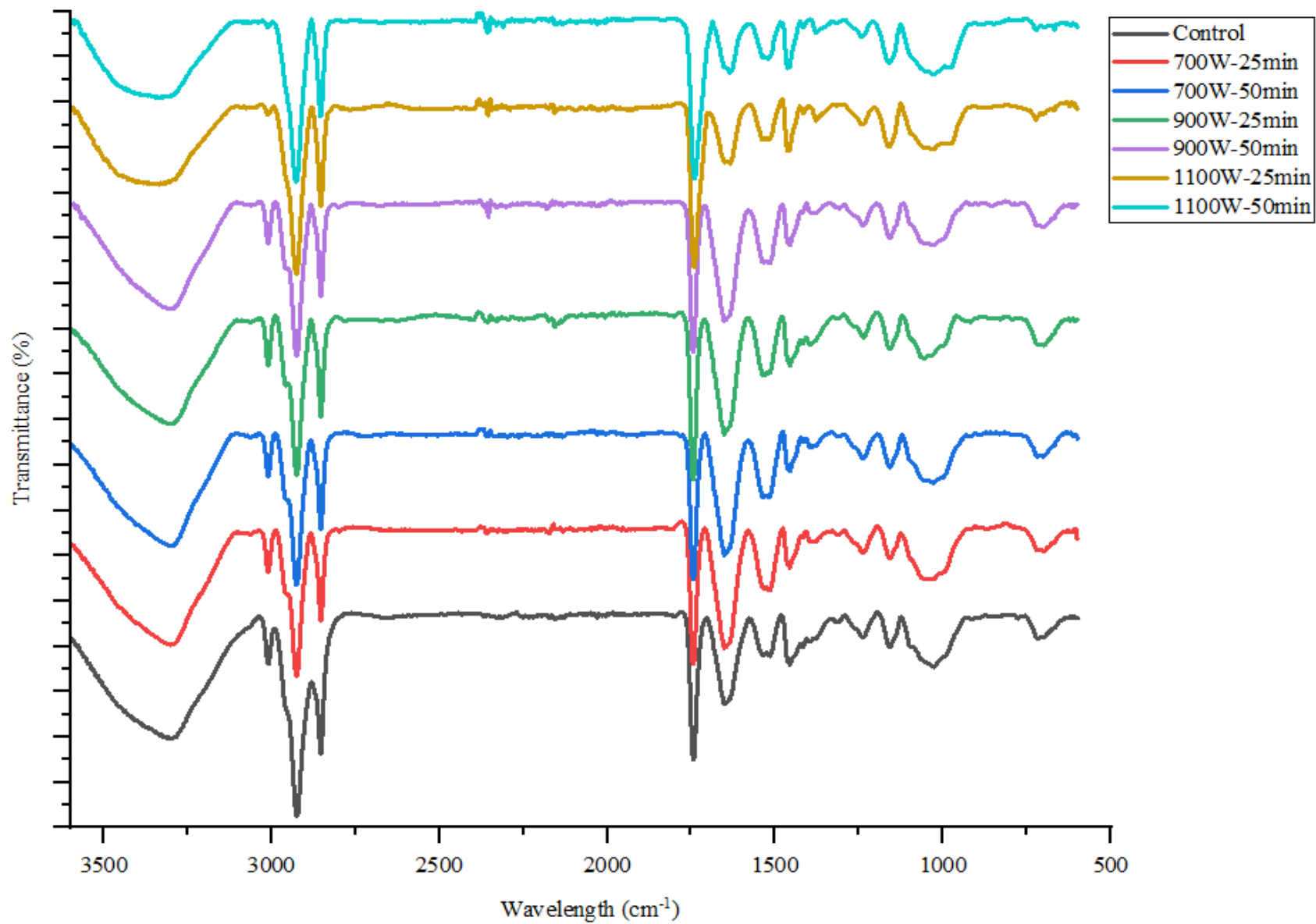


Figure 13. (continued)



**Figure 14.** Single graph for FTIR spectrums of all chia samples

**Table 12.** Specific peaks in the FTIR spectrums given in the literature for chia

Type of Vibration	Band location (cm <sup>-1</sup> )	Reference
OH stretching	3200-3600	(Hatamian et al., 2020)
C=C-H stretching	3009	(García-Salcedo et al., 2018)
C-H stretching	2800-3000	(Hatamian et al., 2020; Noshad et al., 2020)
	2872, 2933, and 2922	(Darwish and El-Sohaimy, 2018)
C=O stretching	1743	(García-Salcedo et al., 2018)
	1748	(Hatamian et al., 2020; Noshad et al., 2020)
	1735	(Darwish and El-Sohaimy, 2018)
Amide I groups	1600	(Hatamian et al., 2020)
-COO- of uronic acids	1618-1915	(Darwish and El-Sohaimy, 2018)
	1422-1595	(Timilsena et al., 2016)
C=O stretching in uronic acids	1500	(García-Salcedo et al., 2018)
	1500-1600	(Noshad et al., 2020)
	1500-1800	(Timilsena et al., 2016)
H-C-H stretching	1400	(García-Salcedo et al., 2018)
Amid III	1246	(Hatamian et al., 2020; Noshad et al., 2020)
C-O-C in the pyranose ring	1155	(Darwish and El-Sohaimy, 2018)
Pyranose ring	1033	(García-Salcedo et al., 2018)
CH <sub>2</sub> with more than 7 carbon atoms	705	(Darwish and El-Sohaimy, 2018)

Overall FTIR results for control and infrared treated chia samples indicated that the intensity of the peak at 3015 cm<sup>-1</sup> decreased in the chia samples infrared treated at 1100W for 25 and 50 min. The decrease in the intensity of this peak was probably due to the oxidation of vinyl groups especially the ones in unsaturated fatty acids.

For control chia sample, a peak was observed at 1636 cm<sup>-1</sup>. The intensity of this peak increased in the samples infrared treated at 700W and 900W for 25 and 50 min as compared to that of control. However, the intensity of this peak decreased in the chia samples infrared treated at 1100W for 25 and 50 min. Hatamian et al. (2020) reported that the change in the peak at 1600 cm<sup>-1</sup> corresponding to Amide I group might indicate the

impact of roasting of chia on amides, amino acids, aldehydes, and esters (Hatamian et al., 2020). The peaks for Amide I group was reported to indicate the secondary protein structure (Tang et al., 2021). A decrease in intensity of the peak at  $725\text{ cm}^{-1}$  was observed for the chia samples infrared treated at 1100W for 25 and 50 min.

## 4.2. Properties of Mucilage

### 4.2.1. Yield of Mucilage Extracted from Infrared Treated Chia Samples

Yield of mucilages extracted from control and infrared treated chia samples are given in Table 13. Mucilage yield of control chia sample was found to be 6.01 g dry mucilage/100g dry chia. In literature, mucilage yield for chia seed was reported as 38 g/kg (dw) (Capitani et al., 2013), 5.81 g/100g for lyophilized mucilage and 4.69 g/100g for mucilage dried at  $50^{\circ}\text{C}$  (Fernandes and Salas-Mellado, 2017), 6.52% (Castejón et al., 2017), 6.97% (Hernández, 2012), and 8% (microwave assisted extraction, Nayani, 2020). Mucilage yield for control chia used in the present study was found to be higher as compared to that reported by Capitani et al. (2013) and Fernandes and Salas-Mellado (2017).

Mucilage yield for infrared treated chia samples were between 6.05-6.99 g dry mucilage/100g dry chia (Table 13). Infrared treatment caused a significant increase in mucilage yield. The increase was significant for all chia samples, except for 1100W-50 min chia sample. There was no significant difference between mucilage yield of control and 1100W-50 min chia sample. Highest mucilage yield (6.99 g dry mucilage/100g dry chia) was obtained for chia seed infrared treated at 1100W-25 min. The mucilage yields for chia samples infrared treated at 700W and 900W were not significantly different.

**Table 13.** Yield of mucilage extracted from infrared treated chia samples\*

Mucilage obtained from chia sample	Mucilage Yield (g dry mucilage/100 g dry chia)
Control	$6.01 \pm 0.318^c$
700W-25 min	$6.39 \pm 0.284^b$
700W-50 min	$6.46 \pm 0.308^b$
900W-25 min	$6.42 \pm 0.304^b$
900W-50 min	$6.68 \pm 0.574^b$
1100W-25 min	$6.99 \pm 0.323^a$
1100W-50 min	$6.05 \pm 0.112^c$

\*Values followed by the same letter are not significantly different ( $\alpha=0.05$ )

Means are based on eleven production replicates.

In the literature, many extraction methods have been used for mucilage extraction from chia seeds. In a study by Hernández (2012), mucilage extraction from chia seeds was



carried out for 2 hours at different seed:water ratios (1:20, 1:30, and 1:40), pH values (4, 6, and 8) and temperatures (4, 40, and 80°C). The mucilage samples were dried at 50°C and highest yield (6.97%) was reported for seed:water ratio (1:40), at 80°C and pH 8. Tavares et al. (2018) used hot and cold mucilage extraction methods. For hot extraction method, seed:water ratio of 1:40, pH 8, 80°C for 2 hours was used. The mucilages were dried at 50°C. For cold extraction method, various seed:water ratio (1:10, 1:20, 1:30, and 1:40), pH 8, 27°C for 2 hours was used. Controlled pressure of 313 kgf/cm<sup>2</sup> was used to separate the mucilage from the seeds and the samples were freeze-dried. The mucilage yield for hot extraction was reported as 6.52%. Higher mucilage yields were obtained for cold extraction method. The cold extraction method did not cause significant differences for various seed:water ratios of 1:20, 1:30, and 1:40 and the mucilage yields were reported as 8.46%, 8.65%, and 8.31%, respectively. As compared to mucilage yield reported by Hernández (2012), in the present study similar mucilage yield (6.01-6.99 g dry mucilage/100 g dry chia) was obtained for seed:water ratio of 1:20 at extraction temperature of 80°C. Tavares et al. (2018) reported higher mucilage yield as compared to the ones found in the present study.

Urbizo-Reyes et al., (2019) also used sonication for separation of mucilage from the chia seeds. It was reported that microjets on chia surface, generated by ultrasound frequencies, targeted the structure (collumnella) and allowed fast and efficient physical separation of mucilage.

#### **4.2.2. Mucilage Composition**

The moisture, protein, and ash contents of the mucilage obtained from the control and infrared treated chia samples are presented in Table 14. The moisture contents of the mucilage samples were between 5.40-5.86%.

The protein content of mucilage samples extracted from control and infrared treated chia samples were between 9.26-10.83% (dw). The protein contents of mucilage samples were found to be lower than those of chia samples (21.33-22.42%, dw (Section 4.1.1)). The protein content of mucilage obtained from control chia sample was 10.83% (dw). Significantly lower protein contents were observed for the mucilage samples obtained from the infrared treated chia samples. In literature, the protein content of mucilage was reported as 4.43 % (Hernández, 2012) and 112g/kg (Capitani et al., 2013).

The ash content of mucilage samples extracted from control and the infrared treated chia samples were found to be between 12.09-12.91% (dw). The ash contents of the mucilage samples were found to be higher than those of the chia samples (4.72-4.80% (dw), Section 4.1.1). Significantly higher ash contents were obtained for the mucilage samples extracted from chia samples infrared treated at 1100W for 25 min and 50 min. In literature, the ash content of mucilage was reported as 8.07 % (Hernández, 2012) and 84 g/kg (Capitani et al., 2013).

**Table 14.** Chemical composition of the mucilage samples\*

Mucilage obtained from chia sample	Moisture (%)	Protein (% dw)	Ash (% dw)
Control	5.80 ± 0.029	10.83 ± 0.131 <sup>a</sup>	12.21 ± 0.043 <sup>bc</sup>
700W-25 min	5.82 ± 0.021	9.92 ± 0.131 <sup>b</sup>	12.09 ± 0.033 <sup>c</sup>
700W-50 min	5.55 ± 0.020	9.80 ± 0.127 <sup>b</sup>	12.11 ± 0.007 <sup>c</sup>
900W-25 min	5.54 ± 0.163	9.26 ± 0.255 <sup>c</sup>	12.18 ± 0.012 <sup>bc</sup>
900W-50 min	5.40 ± 0.040	9.29 ± 0.063 <sup>c</sup>	12.28 ± 0.062 <sup>b</sup>
1100W-25 min	5.42 ± 0.080	9.33 ± 0.129 <sup>c</sup>	12.77 ± 0.104 <sup>a</sup>
1100W-50 min	5.86 ± 0.009	9.29 ± 0.129 <sup>c</sup>	12.91 ± 0.099 <sup>a</sup>

\*Values followed by the same letter in the same column are not significantly different ( $\alpha=0.05$ )  
Values are the means of two replicates.

#### 4.2.3. Color Values of Mucilage Samples

L\*, a\*, b\* color values of the mucilage samples are given in Table 15. L\*, a\*, and b\* color values of mucilage samples were between 49.75-54.67, 2.24-4.24, and 6.64-10.02, respectively.

**Table 15.** Color values of the mucilage samples

Mucilage obtained from chia sample	L*	a*	b*
Control	54.67 ± 0.075	3.38 ± 0.072	9.99 ± 0.006
700W- 25 min	50.37 ± 0.875	2.24 ± 0.231	6.64 ± 0.190
700W-50 min	49.75 ± 0.434	2.40 ± 0.240	7.85 ± 0.488
900W-25 min	50.29 ± 0.447	3.24 ± 0.045	8.56 ± 0.064
900W-50 min	50.01 ± 0.337	3.71 ± 0.130	9.28 ± 0.360
1100W-25 min	49.75 ± 0.558	4.24 ± 0.230	10.02 ± 0.615
1100W-50 min	50.80 ± 0.356	3.90 ± 0.017	8.30 ± 0.344

Values are the means of three replicates.

In a study by Wang et al. (2022), mucilage was extracted from chia by using heat treatment (50°C for 30 or 60 min, 80°C for 30 or 60 min) or ultrasonication together with heat. As compared to mucilage extraction at 50°C, mucilage extraction at 80°C resulted

in lower L\* values (30.80-32.98) of mucilage samples. In the present study, 80°C was used for mucilage extraction and higher L\* values (49.75-54.67) were obtained as compared to the L\* values reported by Wang et al. (2022), indicating light color of mucilage produced in the present study.

#### 4.2.4. Total Dietary Fiber Contents of Mucilage Samples

Total dietary fiber contents (% dw) of the mucilage obtained from the control chia and the chia samples infrared treated at different powers and times are presented in Table 16. The total dietary fiber contents of mucilage samples were between 47.33-63.72% and were higher than the chia samples (34.99 (control) - 37.63%, dw). (Section 4.1.3.)

The total dietary fiber content of the mucilage obtained from control chia was 63.72% and it was not significantly different than that of mucilage obtained from 700W-25 min or 700W-50 min chia sample. Infrared treatment at 900W or 1100W caused significant decreases in total dietary fiber contents of mucilage samples. Lowest total dietary fiber content was observed for the mucilage obtained from 1100W-50 min chia sample. In a study by Coorey et al. (2014), crude fiber content of chia seed gel (mucilage) was reported as 57.84% (dw).

**Table 16.** Total dietary fiber contents of the mucilage samples\*

<b>Mucilage obtained from chia sample</b>	<b>Total Dietary Fiber (% dw)</b>
Control	63.72 ± 0.090 <sup>a</sup>
700W-25 min	63.31 ± 0.330 <sup>a</sup>
700W-50 min	63.59 ± 0.809 <sup>a</sup>
900W-25 min	58.25 ± 0.329 <sup>b</sup>
900W-50 min	56.05 ± 0.568 <sup>c</sup>
1100W-25 min	54.67 ± 0.628 <sup>d</sup>
1100W-50 min	47.33 ± 0.000 <sup>e</sup>

\*Values followed by the same letter are not significantly different ( $\alpha=0.05$ )

Values are the means of two replicates.

It was reported by Oboh et al. (2010) that increased temperature could lead to structural changes in the cell wall architecture, resulting in the breakage of weak bonds between polysaccharide chains and glycosidic linkages in the fiber. The decreased association between fiber molecules and/or depolymerization of the fiber could contribute to solubilization, and result in a decrease in raw fiber content after roasting.

#### 4.2.5. Chemical Structure of Mucilage Samples (determined by ATR-FTIR)

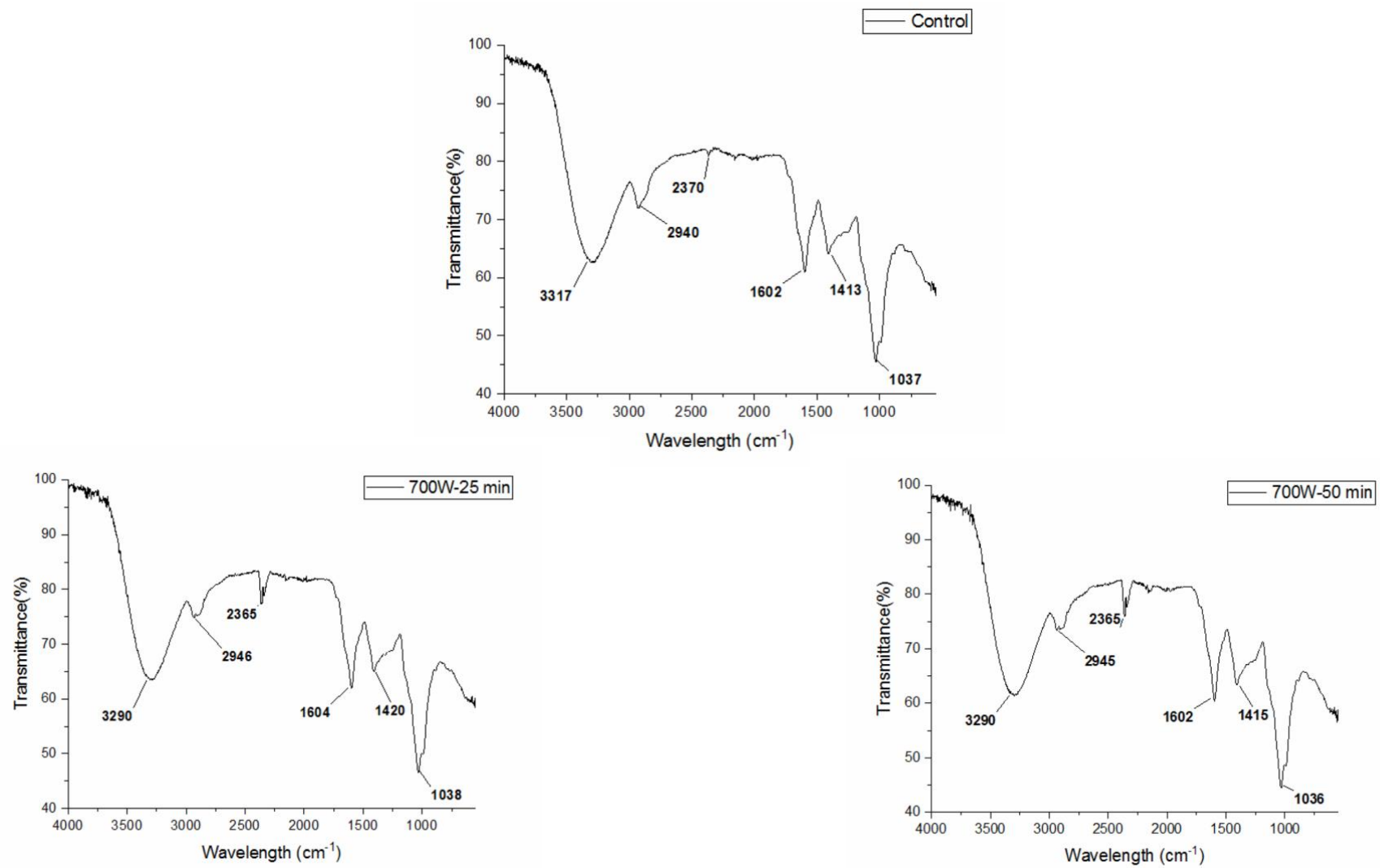
FTIR spectrums of mucilage samples obtained from control and chia samples infrared treated at different powers and times are presented in Figure 15 and Figure 16 (single graph for all mucilage samples). The specific peaks given for mucilage in the literature are summarized in Table 17. Figure 15 and Figure 16 showed that all chia mucilage samples had similar characteristic absorption peaks of chia mucilage given in literature (Table 17).

In the present study, specific peaks for mucilage samples were observed at the wavelengths of  $3290\text{-}3317\text{ cm}^{-1}$ ,  $2940\text{-}2946\text{ cm}^{-1}$ ,  $2363\text{-}2370\text{ cm}^{-1}$ ,  $1602\text{-}1604\text{ cm}^{-1}$ ,  $1413\text{-}1420\text{ cm}^{-1}$ ,  $1250\text{ cm}^{-1}$  or  $1036\text{-}1040\text{ cm}^{-1}$ .

Characteristic absorption peaks of the chia mucilages observed around  $3290\text{-}3317\text{ cm}^{-1}$  could be attributed to OH stretching (Table 17) (Timilsena et al., 2016). The peak observed at  $2940\text{ cm}^{-1}$  could be assigned to the C-H stretching (Darwish and El-Sohaimy, 2018; Ellerbrock et al., 2019; Punia and Dhull, 2019; Silva et al.2022). Stretching of C-H in mucilage samples was previously reported at  $2900\text{ cm}^{-1}$  by Silva et al. (2022),  $2922\text{ cm}^{-1}$  by Ellerbrock et al. (2019),  $2928\text{ cm}^{-1}$  by Punia and Dhull (2019),  $2872$ ,  $2933$ ,  $2922\text{ cm}^{-1}$  by Darwish and El-Sohaimy (2018). The peak observed around  $2363\text{-}2370\text{ cm}^{-1}$  could be attributed to amine salts (Silverstein et al., 1996).

In the present study, a peak was detected at  $1602\text{ cm}^{-1}$ . Silva et al. (2022) reported a peak at  $1630\text{ cm}^{-1}$  and attributed this peak to C=O stretching. However, Timilsena et al. (2016) attributed the peaks around  $1595\text{-}1422\text{ cm}^{-1}$  to -COO- groups in uronic acids. Hernández (2012) reported that peaks around  $1618\text{-}1915\text{ cm}^{-1}$  could be attributed to Amide I groups.

The peak observed around  $1413\text{-}1420\text{ cm}^{-1}$  for chia mucilage samples could be assigned to H-C-H stretching (García-Salcedo et al., 2018) or -COO- groups in uronic acids (Timilsena et al., 2016). The peak detected at  $1250\text{ cm}^{-1}$  could be assigned to C-C stretching (García-Salcedo et al., 2018). The peak detected at  $1036\text{ cm}^{-1}$  for chia mucilage samples could be attributed to the C-O-C stretching of 1→4 glycosidic bonds or C-OH bending or pyranose ring (Darwish and El-Sohaimy, 2018; Ellerbrock et al., 2019; García-Salcedo et al., 2018; Hernández-Nava et al., 2019; Marineli et al., 2014; Silva et al., 2022; Timilsena et al., 2016).



**Figure 15.** FTIR spectrums of mucilage samples extracted from control and infrared treated chia samples

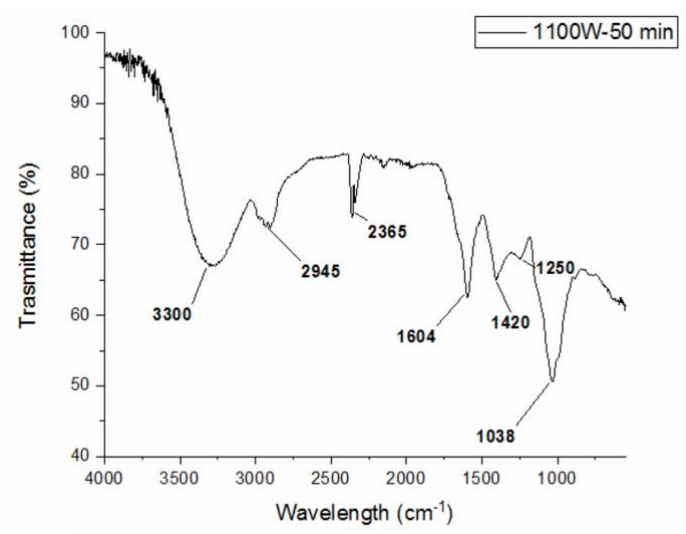
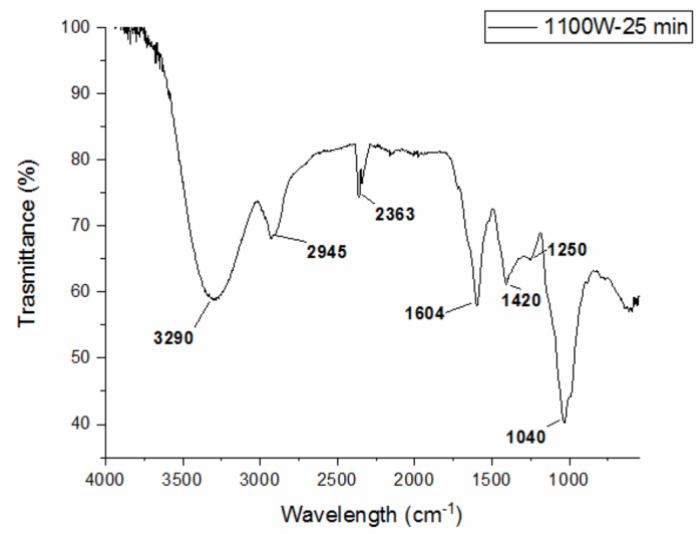
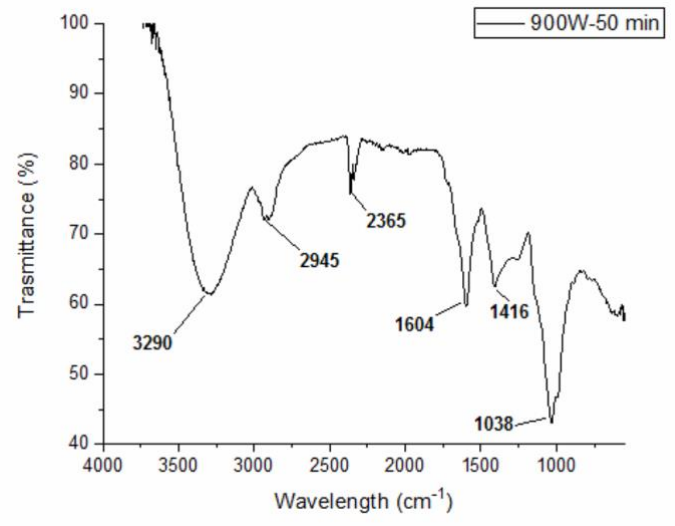
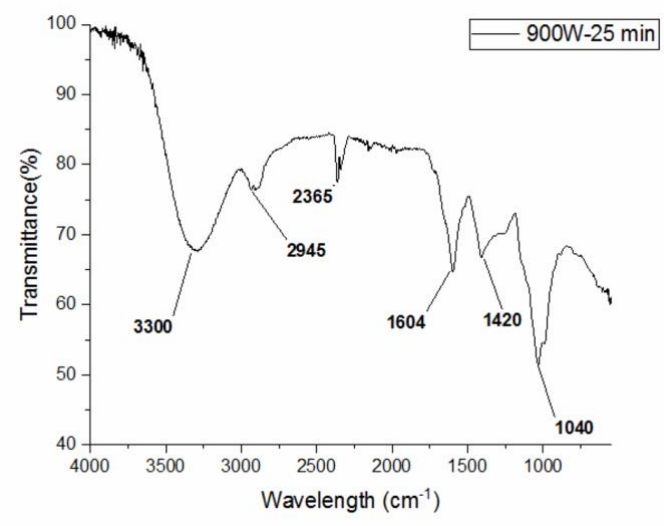
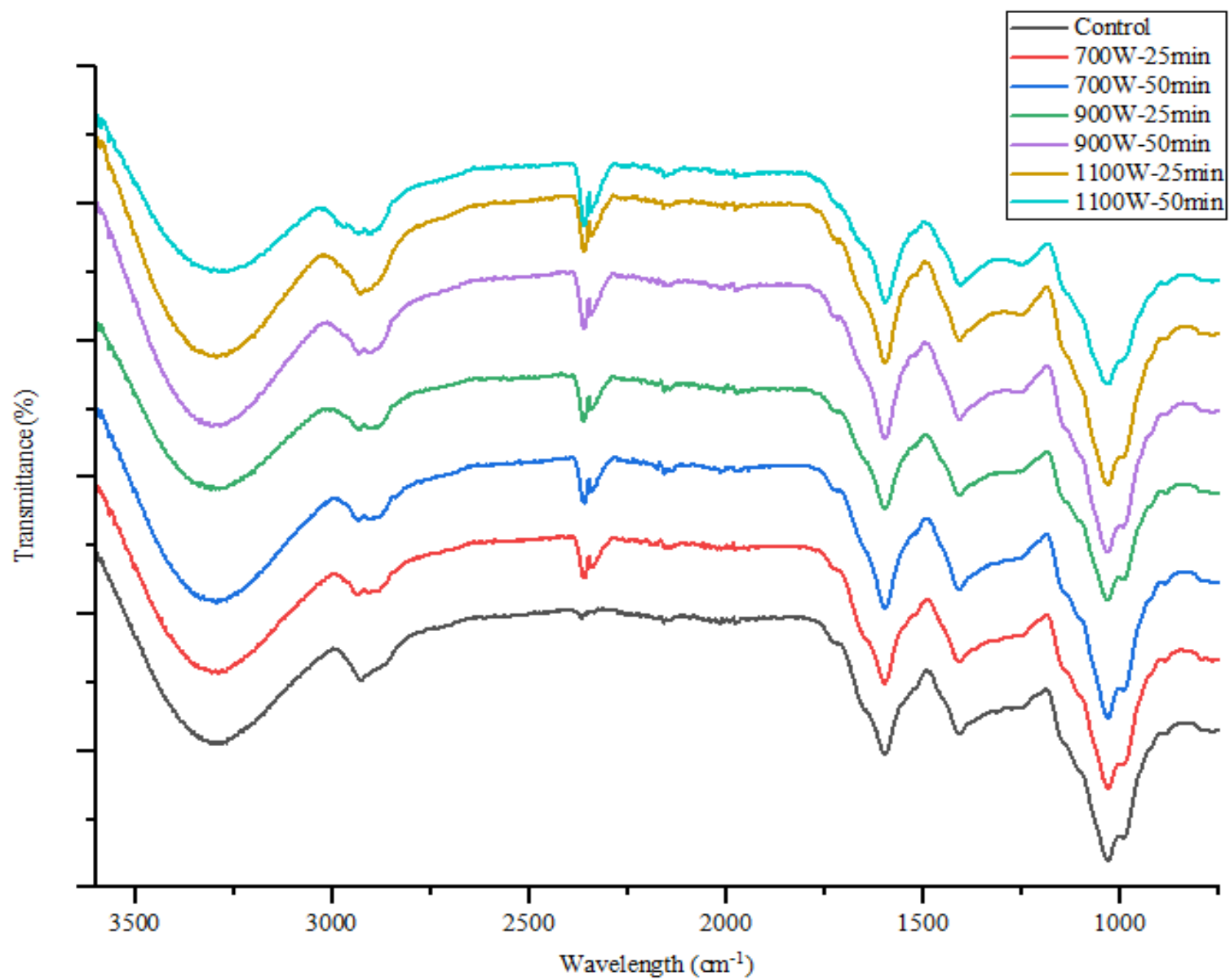


Figure 15. (continued)



**Figure 16.** Single graph for FTIR spectrums of all mucilage samples extracted from control and infrared treated chia samples

**Table 17.** Specific peaks in the FTIR spectrums given in the literature for chia mucilage

Type of Vibration	Band location (cm <sup>-1</sup> )	Reference
OH stretching	3100-3500	(Timilsena et al., 2016)
	3422	(Punia and Dhull, 2019)
	3404	(Ellerbrock et al., 2019)
	3440	(Silva et al., 2022)
C-H stretching	2928	(Punia and Dhull, 2019)
	2872, 2933, and 2922	(Darwish and El-Sohaimy, 2018)
	2922	(Ellerbrock et al., 2019)
	2900	(Silva et al., 2022)
	2800-3000	(Timilsena et al., 2016)
Amine salts	2363-2370	(Silverstein et al., 1996)
-COOH stretching	1800-1500	(Timilsena et al., 2016)
	1743	(García-Salcedo et al., 2018)
C=O stretching	1711	(Ellerbrock et al., 2019)
	1630	(Silva et al., 2022)
Amid-I groups	1618-1915	(Hernández, 2012)
-COO- groups in uronic acids	1595-1422	(Timilsena et al., 2016)
H-C-H stretching	1400	(García-Salcedo et al., 2018)
C-C stretching	1154-1511	(García-Salcedo et al., 2018)
C-O-C stretching of 1→4 glycosidic bonds or C-OH bending	1058	(Darwish and El-Sohaimy, 2018)
	1052	(Ellerbrock et al., 2019)
	1050	(Silva et al., 2022) (Marineli et al., 2014)
	1048	(Timilsena et al., 2016)
	1037	(Hernández-Nava et al., 2019)
Pyranose ring	1033	(García-Salcedo et al., 2018)

Infrared treatment of chia samples resulted in a distinctive peak formation at 2363-2370 cm<sup>-1</sup> which could be attributed to amine salts (Silverstein et al., 1996).

#### 4.2.6. Functional Properties of Mucilage Samples

##### 4.2.6.1. Water and Oil Holding Capacity

The water and oil holding capacity of the mucilage samples obtained from the control or the infrared treated chia samples are given in Table 18. The water holding capacity of mucilage samples were between 1.14-9.70 g water/g dry mucilage and were higher than that of their chia counterparts (except 1100W-50 min) (As given in section 4.1.8.1.). The



water holding capacity of the control chia was 8.44 g water/g dry chia and the infrared treated chia samples were between 1.82-7.61 g water/g dry chia.

The water holding capacity of the mucilage obtained from control chia was 9.70 g water/g dry mucilage and it was not significantly different than that of the mucilage obtained from chia samples infrared treated at 700W or 900W. Water holding capacities of mucilage samples obtained from the chia samples infrared treated at 900W or 1100W for 25 min were not significantly different. Infrared treatment of chia samples at 900W-50min caused a significant decrease in water holding capacity of mucilage. Extremely low water holding capacity was observed for mucilage extracted from 1100W-50 min chia sample. In literature, water holding capacity of mucilage was reported as 5.25 g/g (Darwish et al., 2018) and 15.41 g water/ g FRF (fiber-rich fraction) (Alfredo et al., 2009).

Oil holding capacities of mucilage samples were between 2.01-2.16 g oil/g dry mucilage and were slightly higher than that of their chia counterparts. The oil holding capacity of the control chia was 1.94 g oil/g dry chia and the infrared treated chia samples were between 1.72-1.89 g oil/g dry chia.

Highest (2.16 g oil/g dry mucilage) oil holding capacity was observed for mucilage sample obtained from the control chia. Infrared treatment caused a significant decrease in oil holding capacity of mucilage samples. In literature, oil holding capacity of mucilage was reported as 5.85 g oil/g sample (Darwish et al., 2018) and 2.02 g oil/ g FRF (fiber-rich fraction) (Alfredo et al., 2009).

**Table 18.** Water and oil holding capacity of mucilage samples\*

<b>Mucilage obtained from chia sample</b>	<b>Water Holding capacity (g water/g dry mucilage)</b>	<b>Oil Holding Capacity (g oil/g dry mucilage)</b>
Control	9.70 ± 0.038 <sup>a</sup>	2.16 ± 0.020 <sup>a</sup>
700W-25 min	9.68 ± 0.013 <sup>a</sup>	2.03 ± 0.027 <sup>cd</sup>
700W-50 min	9.68 ± 0.064 <sup>a</sup>	2.09 ± 0.028 <sup>b</sup>
900W-25 min	9.59 ± 0.085 <sup>ab</sup>	2.04 ± 0.020 <sup>cd</sup>
900W-50 min	9.04 ± 0.010 <sup>c</sup>	2.01 ± 0.010 <sup>d</sup>
1100W-25 min	9.52 ± 0.062 <sup>b</sup>	2.07 ± 0.011 <sup>b</sup>
1100W-50 min	1.14 ± 0.075 <sup>d</sup>	2.06 ± 0.024 <sup>bc</sup>

\*Values followed by the same letter in the same column are not significantly different ( $\alpha=0.05$ ) Values are the means of three replicates.

#### 4.2.6.2. Emulsion Activity and Emulsion Stability

The emulsion activity and emulsion stability of the mucilage samples obtained from the control and the infrared treated chia samples are given in Table 19. Emulsion activities of the control mucilage (57.6%) and the mucilage samples (17.3-78.0%) extracted from the infrared treated chia samples were found to be substantially higher than that of their chia counterparts. Emulsion activities of control chia was 20.5% and infrared treated chia samples were between 1.9-22.7%. Emulsion stabilities of the control mucilage (55.5%) and the mucilage samples (16.3-76.2%) extracted from infrared treated chia samples were also substantially higher than that of their chia counterparts. Emulsion stabilities of control chia was 9.9% and infrared treated chia samples were between 1.3-12%.

Infrared treatment of chia samples up to 900W-50 min resulted in an increase in emulsion activity and stability of mucilage samples. Among all mucilage samples, highest (78.0%) emulsion activity and emulsion stability (76.2%) were obtained for the mucilage sample extracted from 900W-50 min chia sample. For chia samples, highest emulsion activity (22.7%) and emulsion stability (12%) values were obtained for the 700W-50 min chia sample.

As the infrared treatment time increased from 25 min to 50 min, the emulsion activities and stabilities increased significantly for the mucilage samples obtained from chia samples infrared treated at 700W and 900W. Lower emulsion activity and stability was obtained for mucilage samples obtained from chia samples infrared treated at 1100W.

**Table 19.** Emulsion activity and emulsion stability of the mucilage samples\*

Mucilage obtained from chia sample	Emulsion Activity (%)	Emulsion Stability (%)
Control	57.6 ± 0.86 <sup>e</sup>	55.5 ± 0.66 <sup>e</sup>
700W-25 min	59.3 ± 0.70 <sup>d</sup>	57.4 ± 0.11 <sup>d</sup>
700W-50 min	64.4 ± 0.16 <sup>c</sup>	63.3 ± 0.88 <sup>c</sup>
900W-25 min	66.9 ± 0.35 <sup>b</sup>	65.3 ± 0.80 <sup>b</sup>
900W-50 min	78.0 ± 0.80 <sup>a</sup>	76.2 ± 0.20 <sup>a</sup>
1100W-25 min	17.9 ± 0.65 <sup>f</sup>	16.3 ± 0.11 <sup>f</sup>
1100W-50 min	17.3 ± 0.67 <sup>f</sup>	16.4 ± 0.67 <sup>f</sup>

\*Values followed by the same letter in the same column are not significantly different ( $\alpha=0.05$ )

Values are the means of three replicates.

In literature, the emulsion activity of mucilage was reported as 53.33 ml/100 ml (solvent fiber-rich fraction) and 44.33 ml/100 ml (pressed fiber-rich fraction) (Capitani et al., 2012) and 61.50% (Coorey et al., 2014). The emulsion stability of mucilage was reported

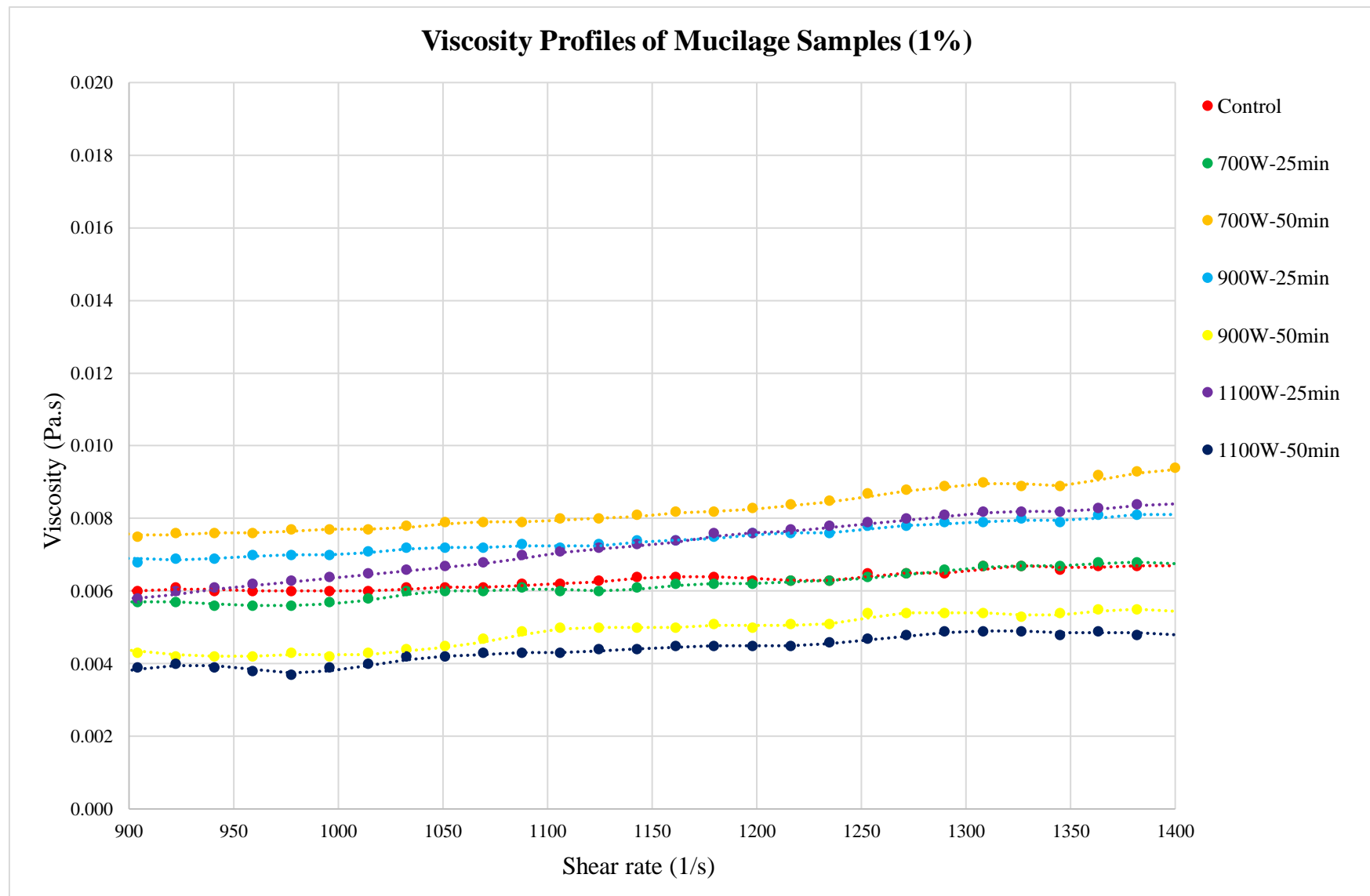
as 57.67 ml/100 ml (solvent fiber-rich fraction) and 34.33 ml/100 ml (pressed fiber-rich fraction) by Capitani et al. (2012) and 69.83% by Coorey et al. (2014).

As compared to the highest emulsion activity value reported in literature (61.50%, Coorey et al., 2014), in the present study, higher emulsion activity values (64.4%, 66.9%, 78%) were observed for mucilage samples obtained from the chia samples infrared treated at 700W-50 min, 900W-25 min, and 900W-50 min. Additionally, the emulsion stability (76.2%) of the mucilage obtained from 900W-50 min chia sample was found to be higher than that of the emulsion stability value (69.83%) reported by Coorey et al. (2014).

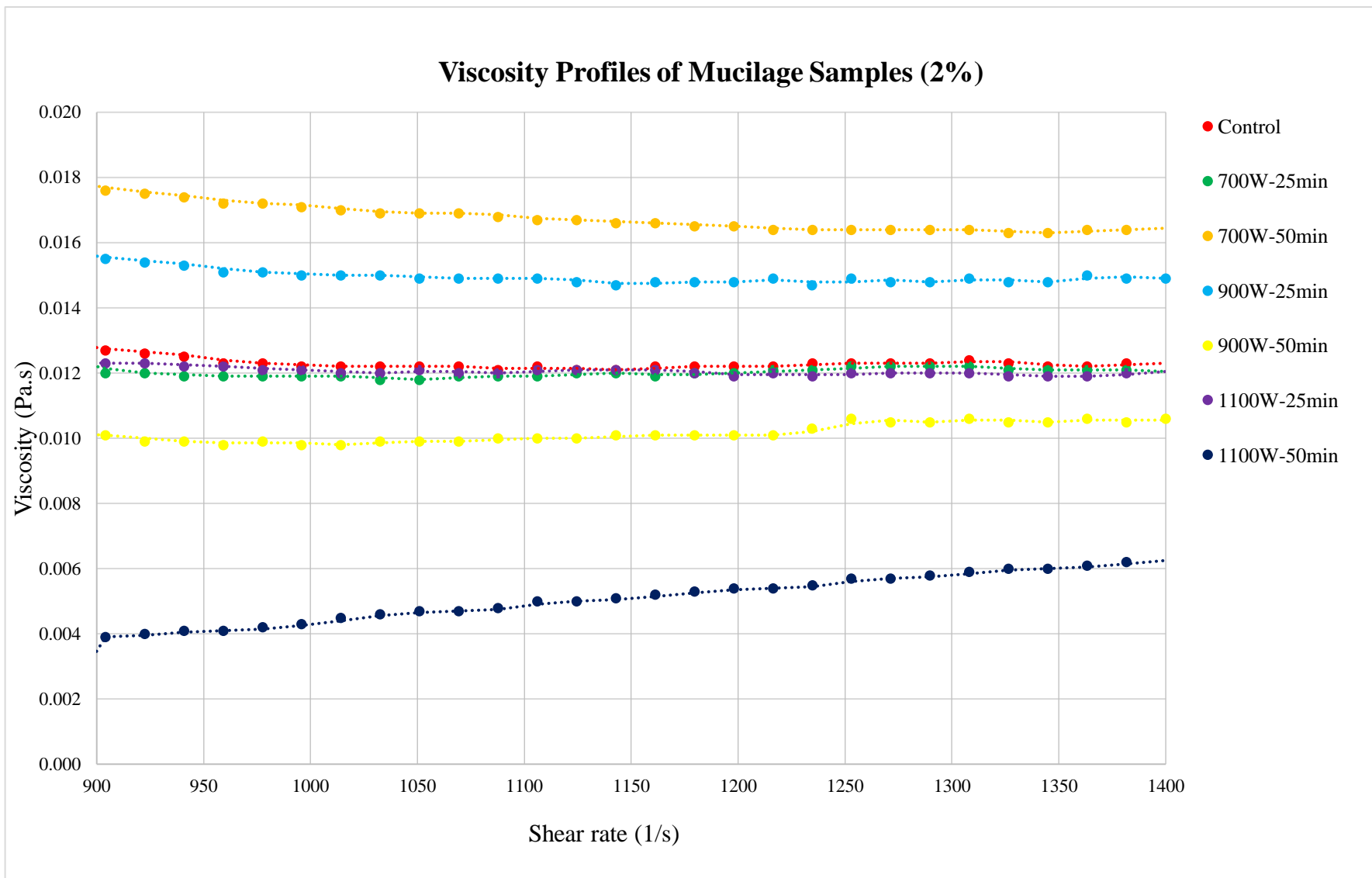
When the emulsion activities of the mucilage samples obtained from the chia samples were evaluated together with chia samples, as compared to their controls significant increase in emulsion activity was observed at 700W-25 min, 700W-50 min, and 900W-25 min. Significant increase in emulsion activity was also observed for mucilage sample obtained from 900W-50 min chia sample while a significant drastic decrease was observed for emulsion activity of 900W-50 min chia sample. When the emulsion stability values were compared with that of control, a significant increase was observed only for chia samples infrared treated at 700W, whereas for mucilage samples obtained from chia samples infrared treated at 700W or 900W.

#### **4.2.6.3. Viscosity Profiles of Mucilage Samples**

The viscosity profiles of mucilage solutions with concentrations of 1% (w/v) and 2% (w/v) are given in Figure 17. As expected, the viscosity increased as the concentration of mucilage increased. For mucilage solutions with a concentration of 1% (w/v) and 2% (w/v), the viscosity of the mucilage samples obtained from 700W-50min and 900W-25min chia samples was higher than that of the mucilage sample obtained from the control. The viscosity of mucilage samples (1% and 2%, w/v) obtained from 700W-25 min chia sample was similar to viscosity of the mucilage samples obtained from control. The viscosity of mucilage sample (1%, w/v) obtained from 1100W-25 min chia sample was higher as compared to the viscosity of control mucilage but at a concentration of 2% (w/v) similar viscosity values were obtained as compared to control. However, infrared treatment of chia samples at 900W-50min and 1100W-50min resulted in lower viscosity values for mucilage samples at the concentrations of 1% (w/v) and 2% (w/v), as compared to that of from the control.



**Figure 17.** Viscosity Profiles of Mucilage Samples (1% and 2%)



**Figure 17.** (continued)

## 5. CONCLUSION

This thesis focuses on the effects of infrared treatment (at different powers and times) on various characteristics of chia seeds. Mucilage was extracted from control and infrared treated chia samples and mucilage properties were determined. The effect of infrared treatment was probably due to the thermal breakdown of cellular compounds and improved extractability.

In the study, chia seeds were infrared treated at 700W, 900W, and 1100W for 25 and 50 minutes. The effects of infrared treatment on protein and ash content, color, total phenolic content, total flavonoid content, phenolic profile (by HPLC), antioxidant activity, total dietary fiber, functional properties (water and oil holding capacity, emulsion activity, emulsion stability), and FTIR spectrum of chia seed were investigated. Mucilage was extracted from control and infrared treated chia samples. The effects of infrared treatment on the yield, protein, ash, color, functional properties (water and oil holding capacity, emulsion activity, emulsion stability, viscosity), and FTIR spectrum of mucilage were investigated.

Infrared treatment at all powers and times caused a significant increase in total phenolic content and antioxidant activity (DPPH and TAC) of chia samples. The highest values were obtained for 1100W-50 min chia sample. Infrared treatment at 700W and 900W caused an increase in total flavonoid content and total dietary fiber content. Infrared treatment caused a significant increase in chlorogenic acid, ferulic acid (except 1100W-50 min) and rutin (not detected for 700W-25 min chia sample). A significant increase was observed in quercetin content of 700W-50 min and 900W-25 min chia samples. Improved extractability through the thermal breakdown of cellular components by the aid of infrared treatment may result in an increase in total phenolic and flavonoid content. This increase contributes to antioxidant activity.

Emulsion activity and emulsion stability significantly increased with an infrared treatment up to 900W-25 min and 700W-50 min, respectively. A significant decrease was observed for other chia samples.

L\* color values decreased from 44.50 (control) to 42.65 (1100W-50 min) by infrared treatment. FTIR spectrums for infrared treated chia samples include all the specific peaks reported in literature for chia. Infrared treatment caused a significant decrease in water

and oil holding capacity. A gradual decrease was observed in water holding capacity and the lowest value was obtained for 1100W-50 min chia sample. A slight decrease was observed for protein content of the chia samples but the decrease was insignificant.

Mucilage was extracted from the control and the infrared treated chia samples. Infrared treatment caused a significant increase in mucilage yield and the highest value (6.99%) was observed for 1100W-25 min chia sample. Protein and ash contents of the mucilages were between 9.26-10.83% (dw) and 12.09-12.91% (dw), respectively. L\* color values of the mucilages decreased from 54.67 (control) to 49.75-50.80 by infrared treatment. Infrared treatment at 700W did not cause a significant change in total dietary fiber content of mucilages while 900W and 1100W caused significant decrease. Infrared treatment caused a decrease in the water holding capacity of mucilages and the decrease was insignificant up to 900W-25 min infrared treatment. A slight decrease was observed for oil holding capacity of mucilages and the decrease was insignificant. Infrared treatment at 700W and 900W caused a significant increase in emulsion activity and stability. But infrared treatment at 1100W caused a drastic and significant decrease in emulsion activity and stability. FTIR spectra for mucilages extracted from the infrared treated chia samples include all the specific peaks reported in literature for mucilage. Infrared treatment at 700W-25min, 700W-50min, 900W-25min and 1100W-25min resulted in similar or higher viscosity values at a concentration of 1% and 2% (w/v), as compared to that of mucilage sample obtained from control. However, infrared treatment of chia samples at 900W-50min and 1100W-50min resulted in lower viscosity values for mucilage samples (1% and 2%, w/v).

Overall results and discussions demonstrated that infrared treatment caused increases in total phenolic content, total flavonoid content (except 1100W), antioxidant activity, chlorogenic acid, rutin, ferulic acid (except 1100W-50 min), quercetin (except 700W-25 min, 900W-50 min and 1100W), emulsion activity (except 900W-50 min and 1100W), emulsion stability (except 900W and 1100W), and total dietary fiber (except 1100W) contents of chia samples. Infrared treatment of chia samples resulted in significant increases in yield, emulsion activity (at 700W and 900W), and emulsion stability (at 700W and 900W) of mucilage samples. However, infrared treatment of chia samples at all powers and times caused a negative effect on water and oil holding capacities of chia and mucilage samples. But the decreases were insignificant for water (up to 900W-25 min) and oil holding capacity of mucilage samples.

Chia has high nutritional value and positive impact on health due to its higher phenolic compounds, essential oils, protein and total dietary fiber as compared to cereals. Infrared treatment of the chia seeds improving the amount of health beneficial constituents of chia in the present study is promising for utilization of chia as a high value raw material in the food industry. Mucilage extracted from chia seeds takes part in many industrial applications (food, chemistry, etc.). Increase in mucilage yield by infrared treatment of chia seeds is also promising. Further studies about the changes in chemical structures of all chia and mucilage samples are needed in order to explain the changes in functional properties arising from the structural changes.



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